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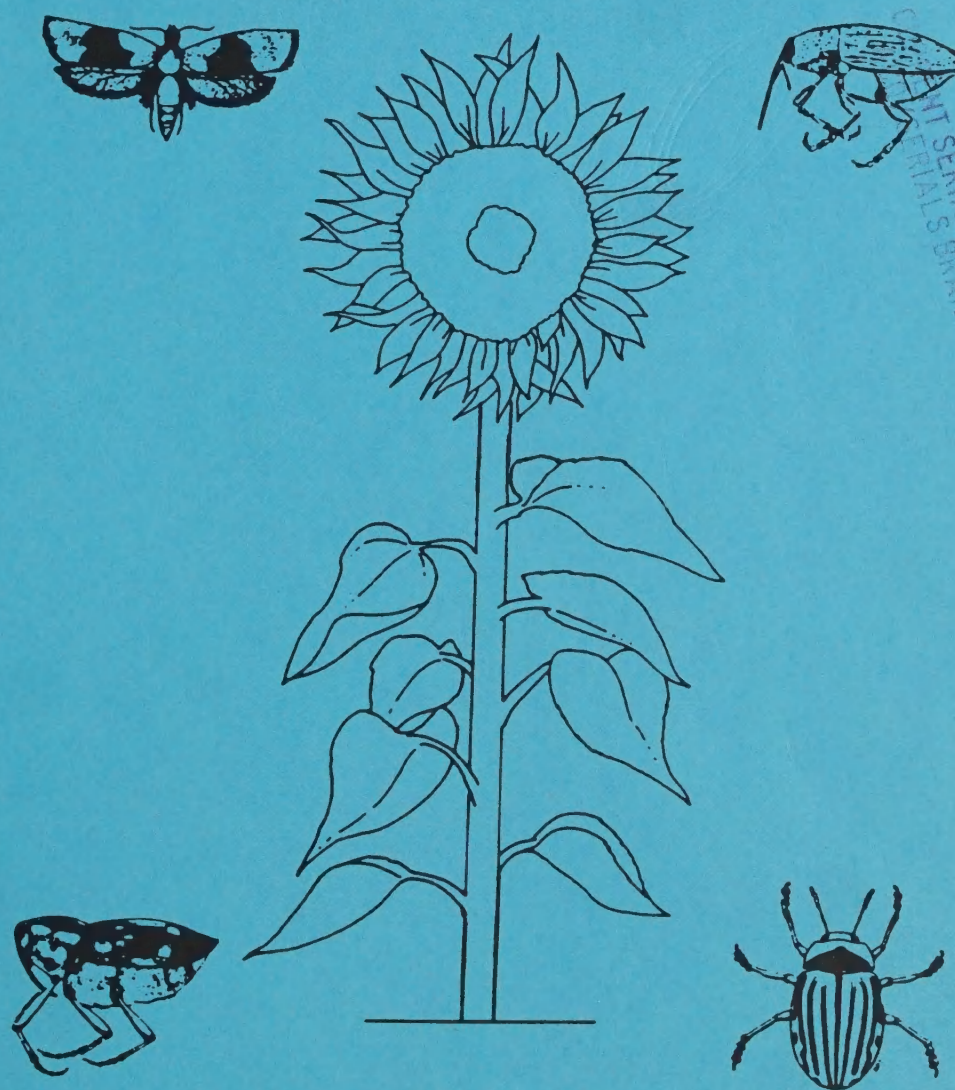
PROCEEDINGS

EIGHTH GREAT PLAINS SUNFLOWER INSECT WORKSHOP



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APRIL 13 - 14, 1994 **FARGO, NORTH DAKOTA**

PROCEEDINGS

EIGHTH GREAT PLAINS SUNFLOWER INSECT WORKSHOP

April 13-14, 1994

USDA, ARS
Northern Crop Science Laboratory
Fargo, North Dakota

Workshop Chair & Proceedings Editor:

Larry D. Charlet
USDA, ARS, Sunflower Insects Research
Northern Crop Science Laboratory
Box 5677, State University Station
Fargo, North Dakota 58105-5677

The Great Plains Sunflower Insect Workshop was developed to foster communication, exchange information, and develop solutions to insect problems of common interest. This volume contains the program, a list of participants and the presentations from the 1994 Workshop. Some of the papers are in summary or abstract form.

The papers in these proceedings are not to be used without the expressed permission of the authors.

Copies of the proceedings are available from the Workshop Chair.

8th Great Plains Sunflower Insect Workshop
13-14 April 1994

U. S. Department of Agriculture, Agricultural Research Service
Northern Crop Science Laboratory
Fargo, North Dakota

Program & Schedule

Wednesday Morning, 13 April

8:00 - 8:30 **Registration**

8:30 - 8:35 **Welcome** - **Don Zimmerman**, *Center Director, Red River Valley Agricultural Research Center, USDA, ARS, Fargo, ND 58105*

Introduction - **Larry Charlet**, *USDA, ARS, Northern Crop Science Laboratory, Fargo, ND 58105.*

8:35 - 10:00 **Presentations**

Vibrational communication in the gray sunflower seed weevil? - **Charles Sawicki**, *Physics Department, North Dakota State University, Fargo, ND 58105.*

Biochemistry and physiology of overwintering in the mature larva of the sunflower stem weevil, *Cylindrocopturus adspersus* (Coleoptera: Curculionidae) in the Northern Great Plains - **Robert Rojas**, *USDA, ARS, Biosciences Research Laboratory, Fargo, ND 58105.*

Indentification and characterization of a bacterial toxin active against the red sunflower seed weevil - **Scott Grayburn**, *USDA, ARS, Northern Crop Science Laboratory, Fargo, ND 58105.*

Insect/plant/microbe interactions: Current research and future plans - **John Barker**, *USDA, ARS, Biosciences Research Laboratory, Fargo, ND 58105.*

Development of strategies for production of adult sunflower seed weevils and banded sunflower moth in the laboratory - **Luming Brewer**, *USDA, ARS, Northern Crop Science Laboratory, Fargo, ND 58105.*

Discussion

10:00 - 10:30 **Break and Refreshments**

Wednesday Morning, 13 April

10:30 - 12:00 **Presentations**

Evaluation of National Plant Germplasm System cultivated and wild type sunflower for resistance to the sunflower moth - **Dick Wilson**, *USDA, ARS, Plant Introduction Station, Iowa State University, Ames, IA 50011.*

Screening sunflower germplasm for resistance to the red sunflower seed weevil - **Huihua Gao**, *Department of Entomology, North Dakota State University, Fargo, ND 58105.*

Trap cropping as an insect managment tool - **W. Gene Schmidt**, *Department of Entomology, North Dakota State University, Fargo, ND 58105.*

Trap cropping to manage the red sunflower seed weevil - **Gary Brewer**, *Department of Entomology, North Dakota State University, Fargo, ND 58105.*

Discussion

12:00 - 1:30 **Lunch**

Wednesday Afternoon, 13 April

1:30 - 2:30 **Presentations**

North Central Regional Plant Introduction Station's sunflower collection - **Mary Brothers**, *USDA, ARS, North Central Regional Plant Introduction Station (NC-7), Iowa State University, Ames, IA 50011.*

Preparing for banded sunflower moth activity in 1994 and beyond - **Phil Glogoza**, *Extension Service, Box 5346, Department of Entomology, North Dakota State University, Fargo, ND 58105.*

Discussion

2:30 - 3:00 **Break and Refreshments**

3:00 - 4:00 **Presentations**

The use of nematodes as biological control agents and their potential for the control of sunflower insect pests - **Chris Wozniak**, *USDA, ARS, Northern Crop Science Laboratory, Fargo, ND 58105.*

Wednesday Afternoon, 13 April

3:00 - 4:00 **Presentations (cont.)**

Natural control of sunflower insect pests by indigenous parasitoids - **Larry Charlet**, *USDA, ARS, Northern Crop Science Laboratory, Fargo, ND 58105.*

Discussion

Thursday Morning, 14 April

8:30 - 10:00 **Presentations**

Feeding behavior of the red and gray sunflower seed weevils - **Rico Rana**, *Department of Entomology, North Dakota State University, Fargo, ND 58105.*

Economic injury levels for the red sunflower seed weevil (Coleoptera: Curculionidae) - **Chengwang Peng**, *Department of Entomology, North Dakota State University, Fargo, ND 58105.*

Comparison of lab-determined deterrence with field cage deterrence to feeding by the sunflower beetle - **Craig Roseland and Teri Grosz**, *Department of Entomology, North Dakota State University, Fargo, ND 58105.*

Patterns of phenylpropanoid expression and deterrence to leaf feeding - **Craig Roseland and Teri Grosz**, *Department of Entomology, North Dakota State University, Fargo, ND 58105.*

Discussion

10:00 - 10:30 **Break and Refreshments**

10:30 - 11:30 **Presentations**

Sunflower insect pest situation during 1992 and 1993 and prospects for 1994:

North Dakota - **Dave Nelson**, *North Dakota Department of Agriculture, North Dakota State University, Fargo, ND 58105.*
South Dakota - **Murdick McLeod**, *Extension Service, South Dakota State University, Brookings, SD 57007.*
Colorado - **Sue Blodgett**, *Department of Entomology, Colorado State University, Fort Collins, CO 80526.*

Discussion

Thursday Morning, 14 April

11:30 - 12:00 **Research plans for 1994**

Conclusion of Workshop - comments, questions, future plans, etc.

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13-14 April 1994
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Vibrational Communication in the Gray Sunflower Seed Weevil?

By Charles A. Sawicki and Fen Chen
Physics Department, North Dakota State University
Fargo North Dakota 58105

SUMMARY

Substrate vibrations produced by the male gray sunflower seed weevil (GSSW) which may be used for communication were studied and digitally recorded in this work. These vibrations are created by male GSSWs in conjunction with production of a patterned ultrasonic song which appears to be related to mating. In these experiments, a relatively inexpensive method was used to detect these vibrations with amplitudes of less than 1 Å. Substrate vibrations were sensed with a one inch diameter piezoelectric disks (piezo disks) similar to those used in solid state buzzers. Since the piezoelectric effect is reversible, piezo disks were also used to generate artificial substrate vibrations in sunflower stems.

In this experiment, ultrasonic GSSW sounds and the associated substrate vibrations were digitally recorded and signal processed to remove background noise. Our studies show that vibrational signals produced by the GSSW have much lower frequencies (1 to 6 kHz) as compared to the ultrasonic song, which is most intense in the range 10 to 35 kHz. The sunflower plant was found to act as a filter that passes frequencies of 5 ± 1 kHz most efficiently. This filtering action appears to act to increase the range over which substrate vibrational signals stand out above background noise.

An arbitrary function generator was used to create analog vibrational signals from digitally recorded signals produced by male GSSWs. These analog signals were stored on cassette tape, played back to a piezo disk plugged into sunflower plants and detected with another piezo disk plugged into the same plant. The wet summer of 1993 greatly reduced the weevil population and we were not able to play these artificial vibrational songs back to female weevils to see if their behavior would be affected.

INTRODUCTION

The gray sunflower seed weevil, *Smicronyx sordidus* is a pest that attacks cultivated sunflower, *Helianthus annuus* L. reducing achene weight and oil production. Both the gray and red sunflower seed weevils produce sounds with a similar file and scraper type of mechanism (Hyder and Oseto, 1989).

The male gray sunflower seed weevil (GSSW) produces patterened ultrasonic sounds when attempting to mate with a female. During production of this song, other females are observed to become much more active and race around on the bud. Playing back the male ultrasonic sound was observed to have no affect on the behavior of females. In the case of other small insects, substrate vibrations, rather than sound traveling through the air, have been found to be the primary mode of communication. This work represents an attempt to detect substrate vibrations produced by the GSSW.

METHODS AND EQUIPMENT

Gray sunflower seed weevils, *Smicronyx sordidus* LeConte were hatched from larvae collected from harvested sunflower heads. In the laboratory, weevils were kept on freshly cut sunflower heads and used in studies within three days.

Weevils were sexed visually using rostrum or proboscis length forward of antennal attachment.

Digital Vibration and Sound Recording and Playback System

Digital vibration and sound recordings were made in a sound insulating chamber. Weevils were placed on freshly cut 1.5 to 3 cm diameter *Helianthus Annuus* sunflower buds in the chamber. A separate microphone and amplifier (Bionic Ear, Silver Creek Industries, Manitowoc, Wisconsin) were used to monitor sounds while data was collected simultaneously using an ultrasonic microphone (SM2, Ultasound Advice, London, England) and a one inch diameter piezo disk (Murata part number 7BB-27-4) plugged into the sunflower bud. An

optical magnifier mounted in the chamber wall (catalog No. R36934, Edmund Scientific, Barrington, New Jersey) was used to observe weevil behavior while data was collected. Light inside the chamber was provided by a 15 Watt fluorescent lamp. Temperature in the chamber was $28 \pm 3^{\circ}$ C.

Signals from the piezo detector and microphone were amplified by wide band, low-noise amplifiers (Model SR560, Stanford Research Systems, Sunnyvale, California). Amplified signals were digitally recorded with a transient recorder (Model R1200M, Rapid Systems, Seattle, Washington) interfaced to a microcomputer. Figure 1 presents a schematic diagram of the data collection system. The arbitrary function generator was used to convert digital, signal processed data back into analog form so that sounds and vibrations can be played back to the GSSWs in a controlled manner to study their response.

Signal Processing of Sound Data

386-MatLab was used for signal processing and plotting of all data. A preamplifier/filter represented as **PA FILTER** in figure 1 included 100X amplification and filtering of the small vibrational signals produced by the piezo disk. Filtering was carried out with a 4 pole Chebyshev highpass filter with cutoff frequency set to 300 Hz. Sound data sets were digitally filtered with a Chebyshev type I highpass digital filter of order 16 with a cutoff frequency of 600 Hz to remove low frequency noise which was not sufficiently attenuated by the sound insulating chamber.

Discrete Fourier transforms of sound data sets were calculated using Matlab's fast Fourier transform algorithm. Spectral densities plotted in this work were calculated from the Fourier transforms as sound intensity per Hertz versus frequency. Plots of intensity per Hertz versus frequency show how sound intensity is distributed according to sound frequency in recorded signals.

Results and Discussion

Figure 2 represents calibration measurements for the piezo disk used to detect substrate vibrations. The sound intensity in W/m^2 produced on the axis of

the piezo disk is plotted against the amplitude of the 5000 Hz sinusoidal voltage used to drive the disk. For an ideal piezoelectric material the amplitude of surface motion should be directly proportional to applied voltage. The upper solid, straight line which fits the data is described by the equation $6.99 \cdot 10^{-4} \cdot (\text{Volts})^2$ while the lower straight line is given by $2.87 \cdot 10^{-4} \cdot (\text{Volts})^2$. Since the sound intensity is proportional to the square of the amplitude of surface motion, figure 2 shows that voltage and displacement are, as expected, linearly related for the piezo disks used in this work.

Figure 3 presents a measurement of the relative transmission of substrate vibrations by 0.1 m of sunflower stalk. Most efficient transmission takes place in the range 5000 ± 1000 Hz.

Figure 4 presents a simultaneous recording of a male GSSW ultrasonic song (A) and the associated substrate vibrations (A). Each tooth impact that generates an impulse of ultrasound also generates an impulse of substrate vibrations in the sunflower. In part B, 1 vertical unit represents a bending of the piezo disk by approximately 0.01 Å.

Figure 5 presents the fourth to the last impulses of figure 4 on an expanded time scale. The substrate vibration (B) is seem to be much lower in frequency than the ultrasonic sound impulse (A). Figure 6 gives the frequency content of the impulses shown in figure 5. Part A is for the ultrasonic sound impulse while part B is for the substrate vibration impulse.

Figure 7 presents the result of an experiment where substrate vibrational waves were generated in a sunflower stalk by applying the voltage signal in part A to a piezo disk plugged into the stalk. A second piezo disk 0.04 m away detected the signal shown in part B. Although the signal in B is about 10^7 times less intense than that in A the amplitude patterning of this signal is much clearer than that in part A. In particular, note the small impulses at the start of the song which are much more clearly seen in B than in A.

In general, insects use amplitude patterning rather than frequency content to carry their communication signals. Filtering by the sunflower appears to make this amplitude patterning clearer. Figure 8 represents the fourth to the last impulses from figure 7. Part A shows the applied voltage impulse while part B shows the corresponding received vibrational impulse. Figure 9 gives the frequency distributions for the data of figure 8. The applied signal has significant frequency components in the range from 750 to 1500 Hz while the received impulse, after filtering through the plant, only has frequencies close to 5000 Hz.

Future Work

Attempts to influence weevil behavior using the male's ultrasonic song were unsuccessful. We will next try playing back simulated vibrational signals (like that shown in figure 7) to female weevils on sunflower buds to see if their behavior can be altered.

ACKNOWLEDGEMENT

Thanks to Dr. Gary Brewer, Dr. Larry Charlet Dr. Craig Roseland, and Mrs. Luming Brewer for supplying gray sunflower seed weevils used in this study.

REFERENCE

Hyder, D. E. and Oseto, C.Y. (1989) Structure of the stridulatory apparatus and analysis of the sound produced by *Smicronyx fulvus* and *Smicronyx sodidus* J. Morph. 201:69-84.

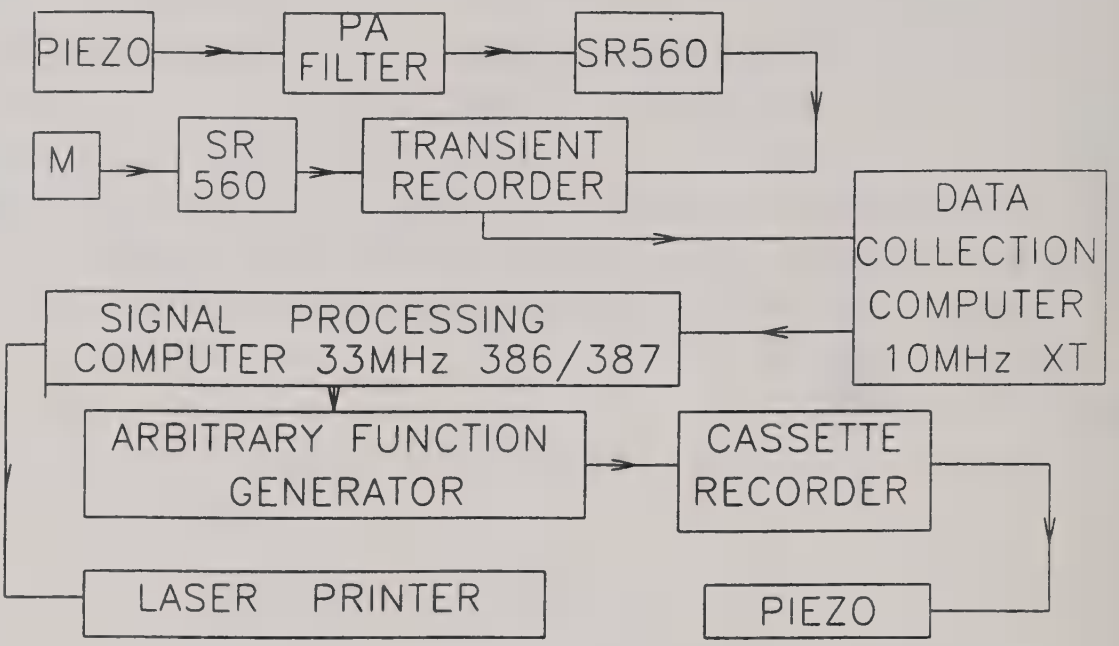

Figure 1

Schematic diagram of system used to make simultaneous recordings of sounds and substrate vibrations produced by male gray sunflower seed weevils.

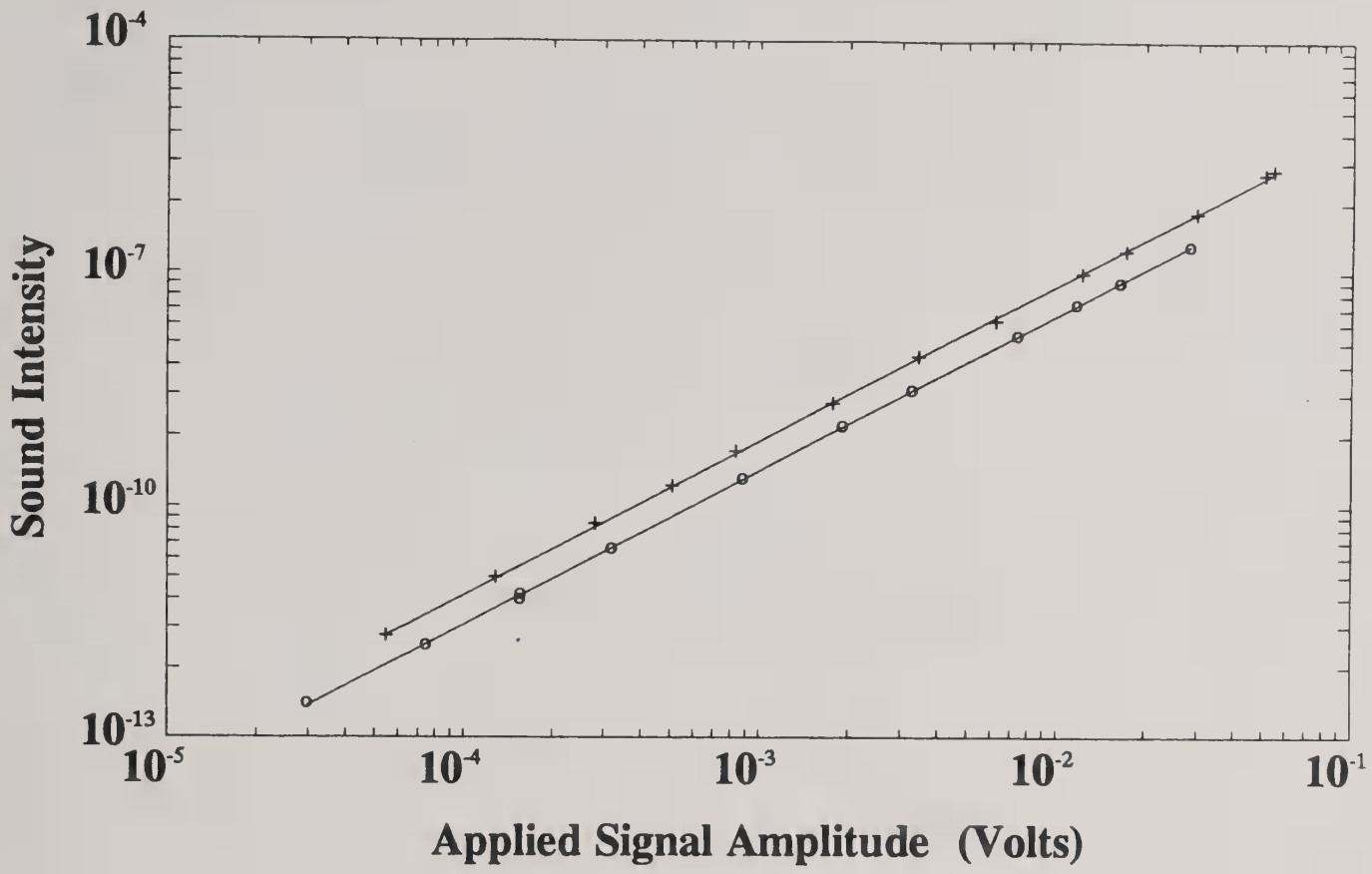

Figure 2

Calibration of piezo disk vibration detector. The amplitude of the 5000 Hz signal applied to the piezo disk is plotted on the horizontal axis while recorded sound intensity is plotted on the vertical axis for two separations between the piezo disk and the microphone. Data represented by the pluses were recorded for a separation of 0.016 meters while the circles represents data recorded at 0.026 meters.

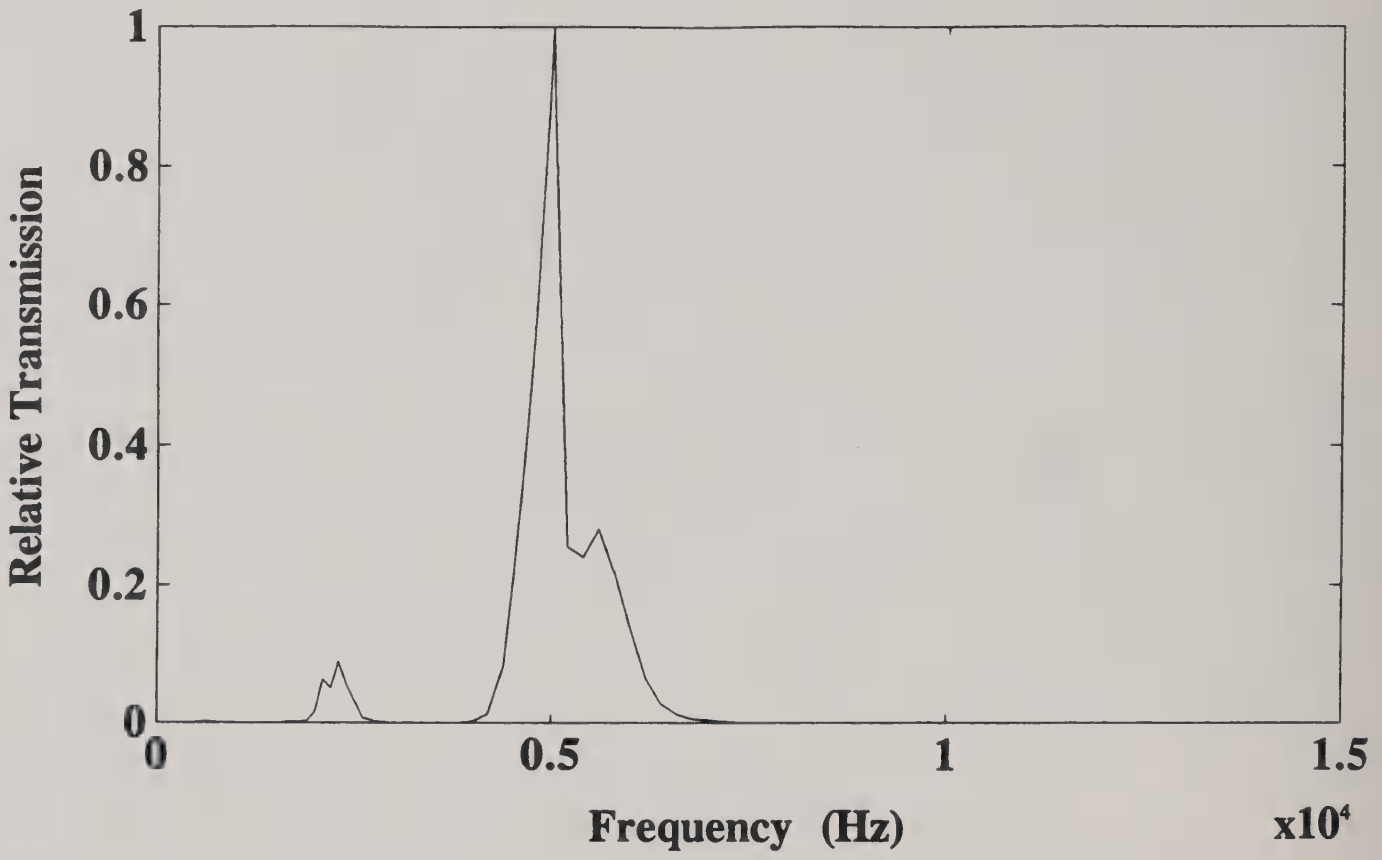

Figure 3

Relative transmission versus frequency of 0.1 meter of sunflower stalk. Piezo disks were used both to generate and detect sinusoidal signals. The relative transmission was calculated from the ratio of the square of the voltage amplitude of the received to the generating signal.

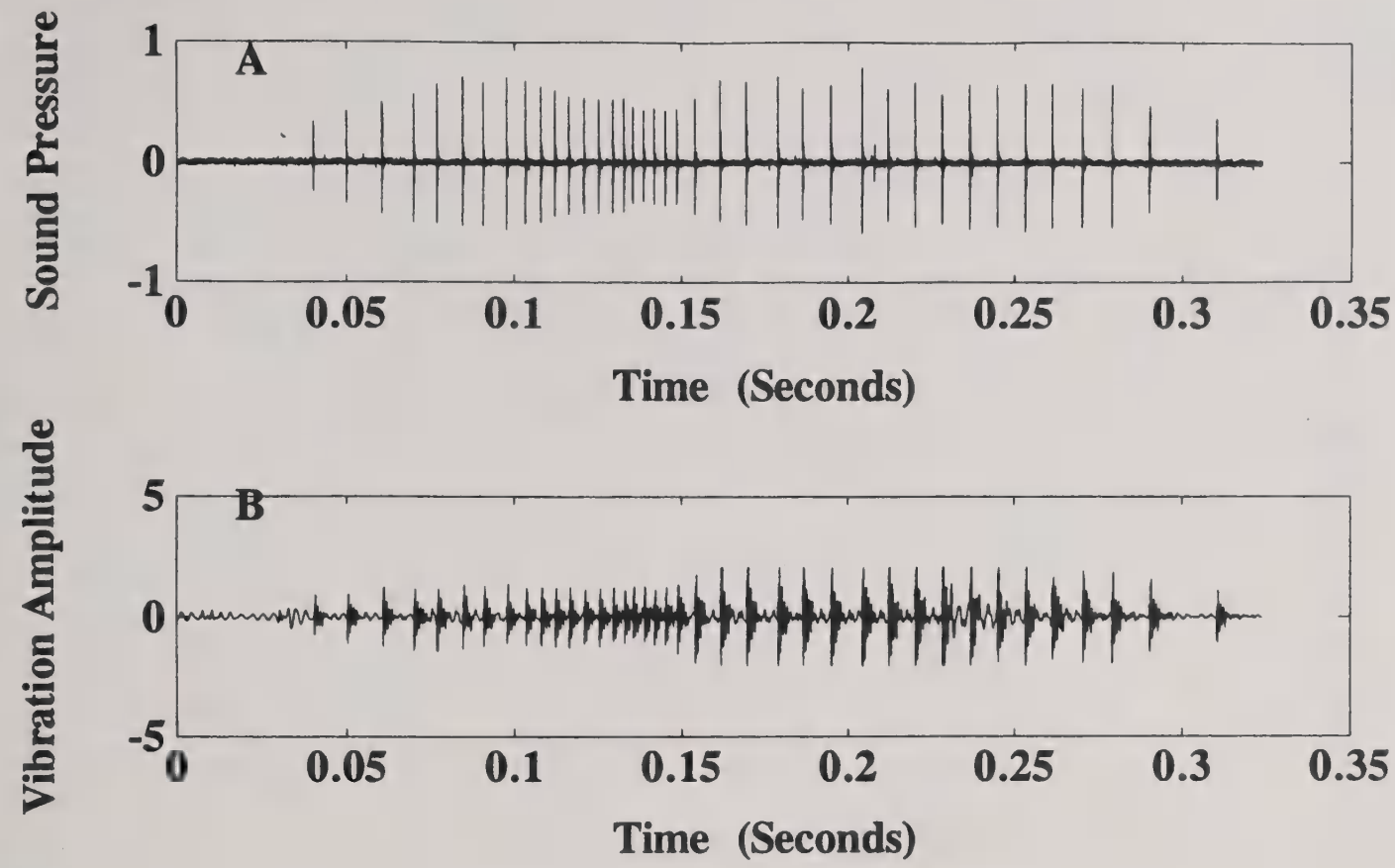

Figure 4

Simultaneous recording of patterned ultrasonic song (A) and substrate vibrational signal (B) produced by a male gray sunflower seed weevil. Data was recorded at 200,000 points per second.

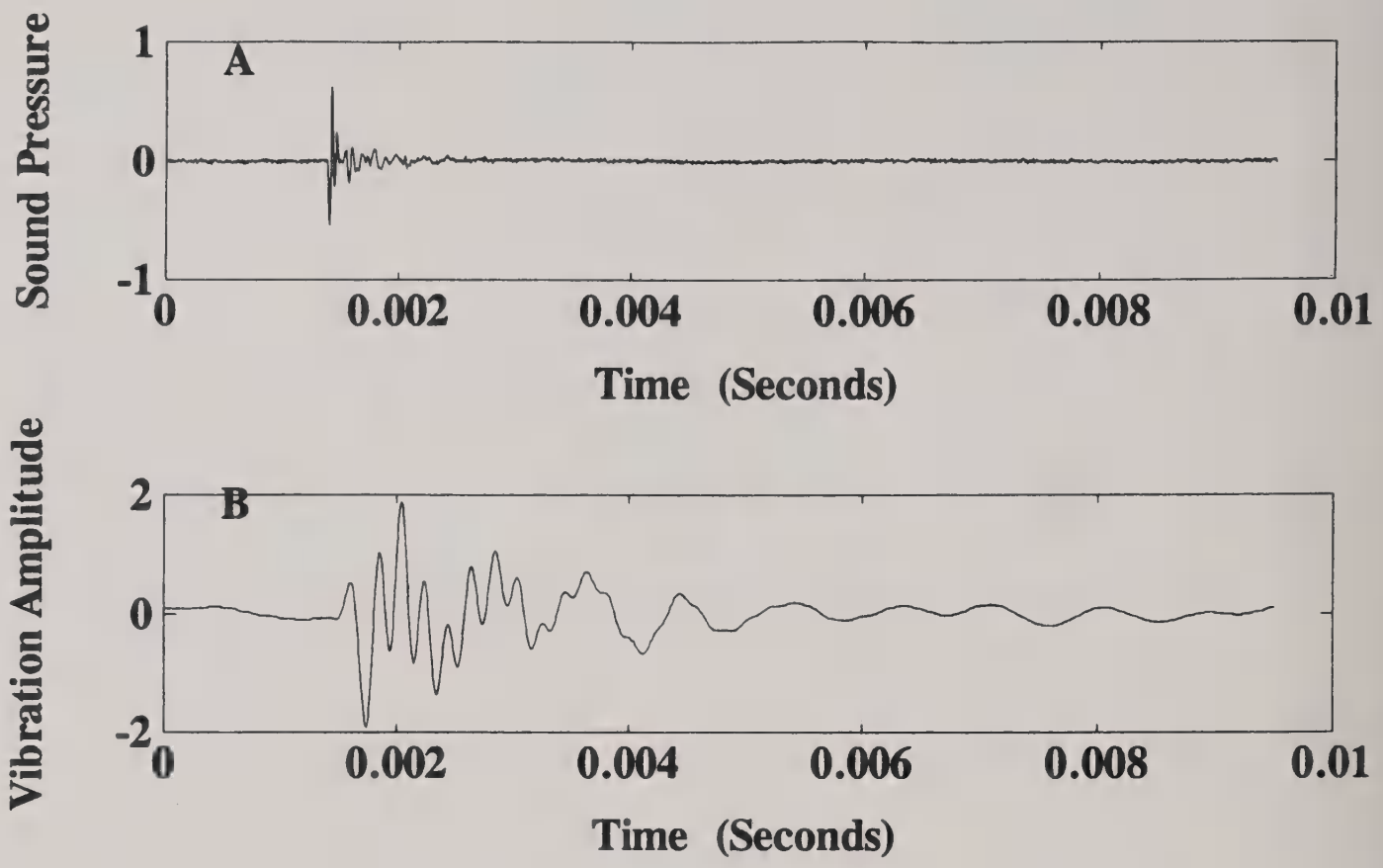

Figure 5

Fourth to the last impulses from figure 4. Part A is the ultrasonic impulse while part B is the corresponding substrate vibration impulse.

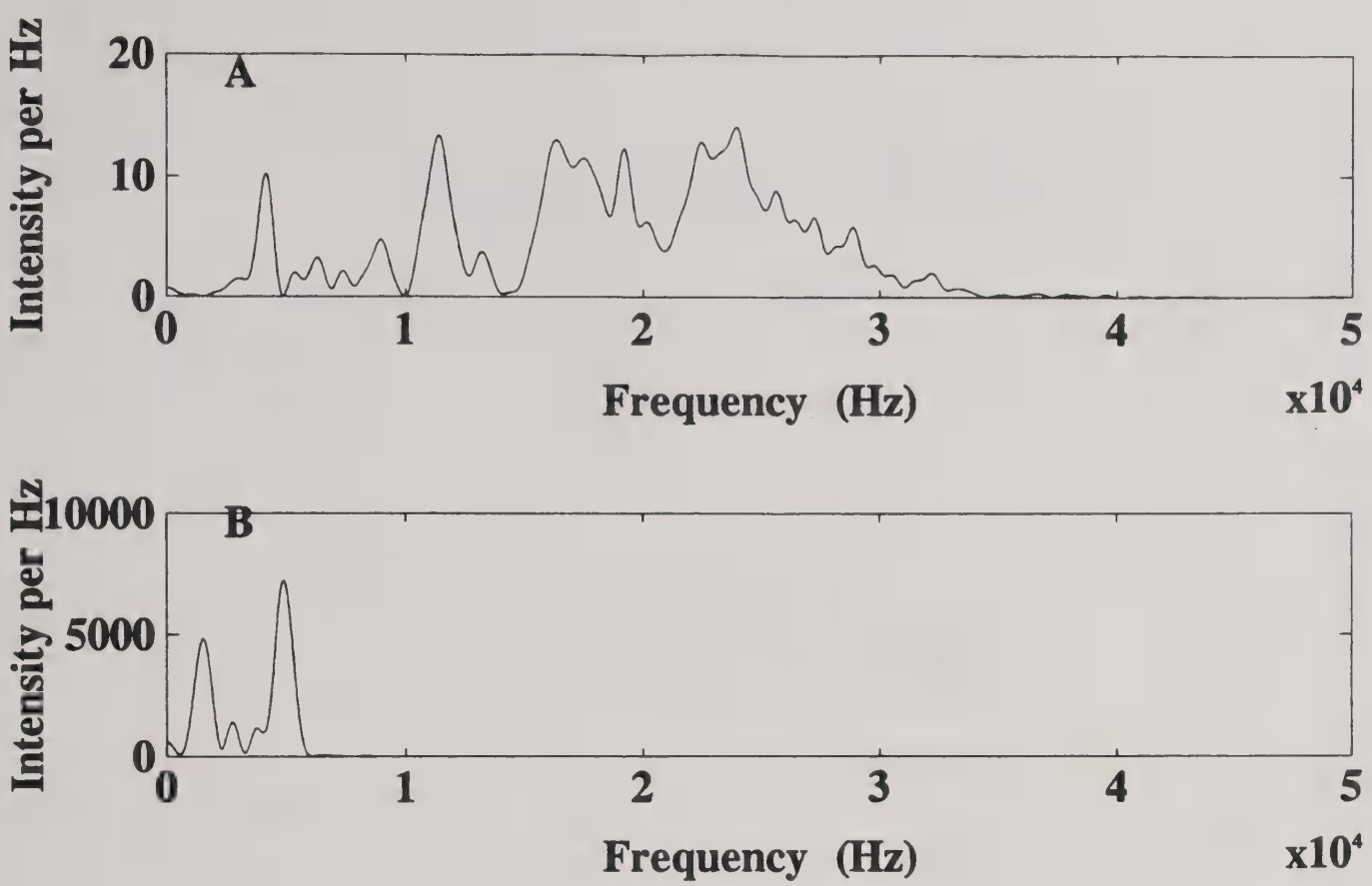

Figure 6

Frequency content of the impulses of figure 5. Part A is spectrum of the ultrasonic sound impulse while B is the spectrum of the corresponding substrate vibration impulse.

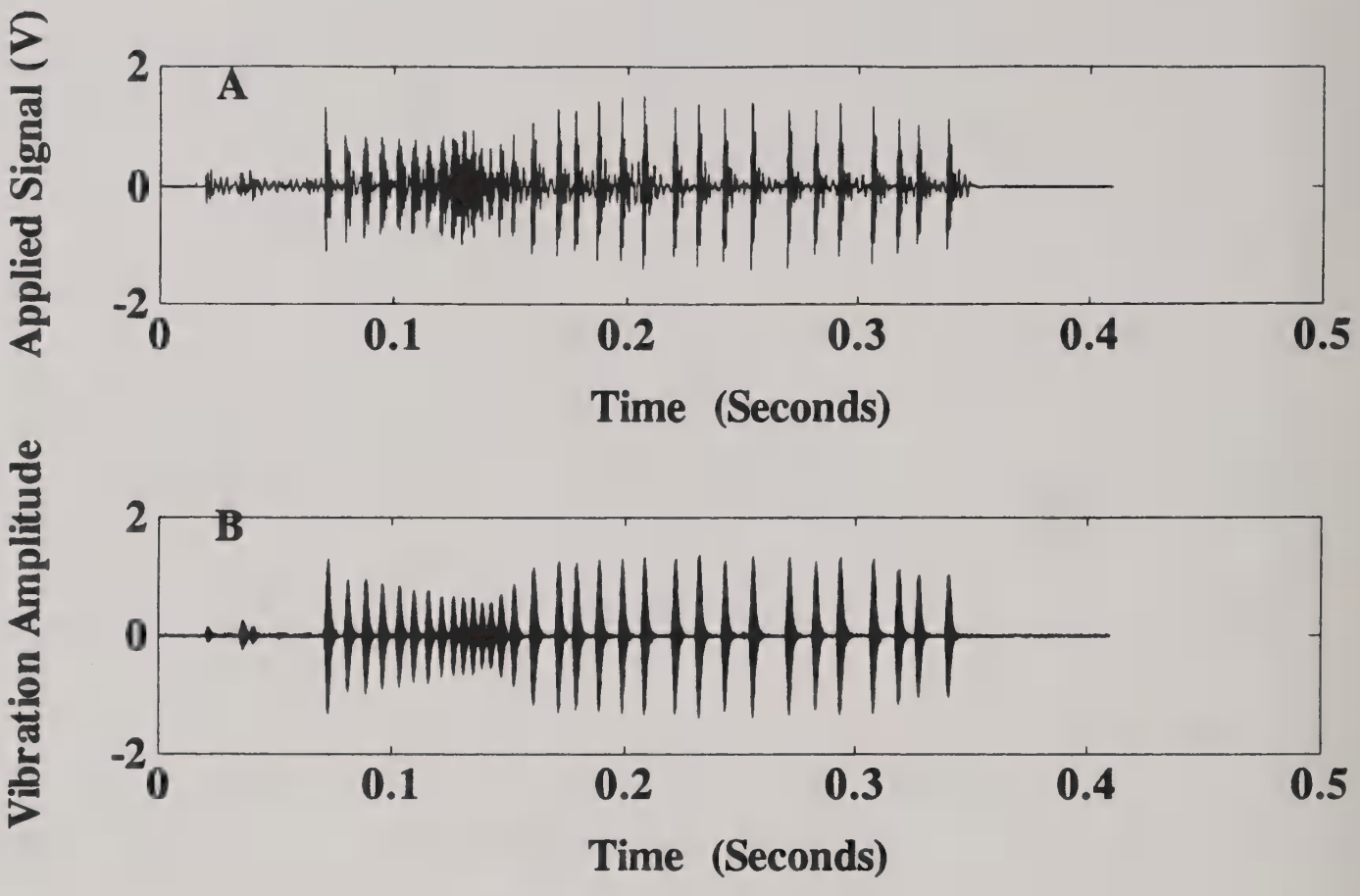

Figure 7

Recorded weevil substrate vibrational song played back to a piezo disk plugged into a sunflower stem (A). Substrate vibration (B) detected by a second piezo disk plugged into the stem 0.04 meters away.

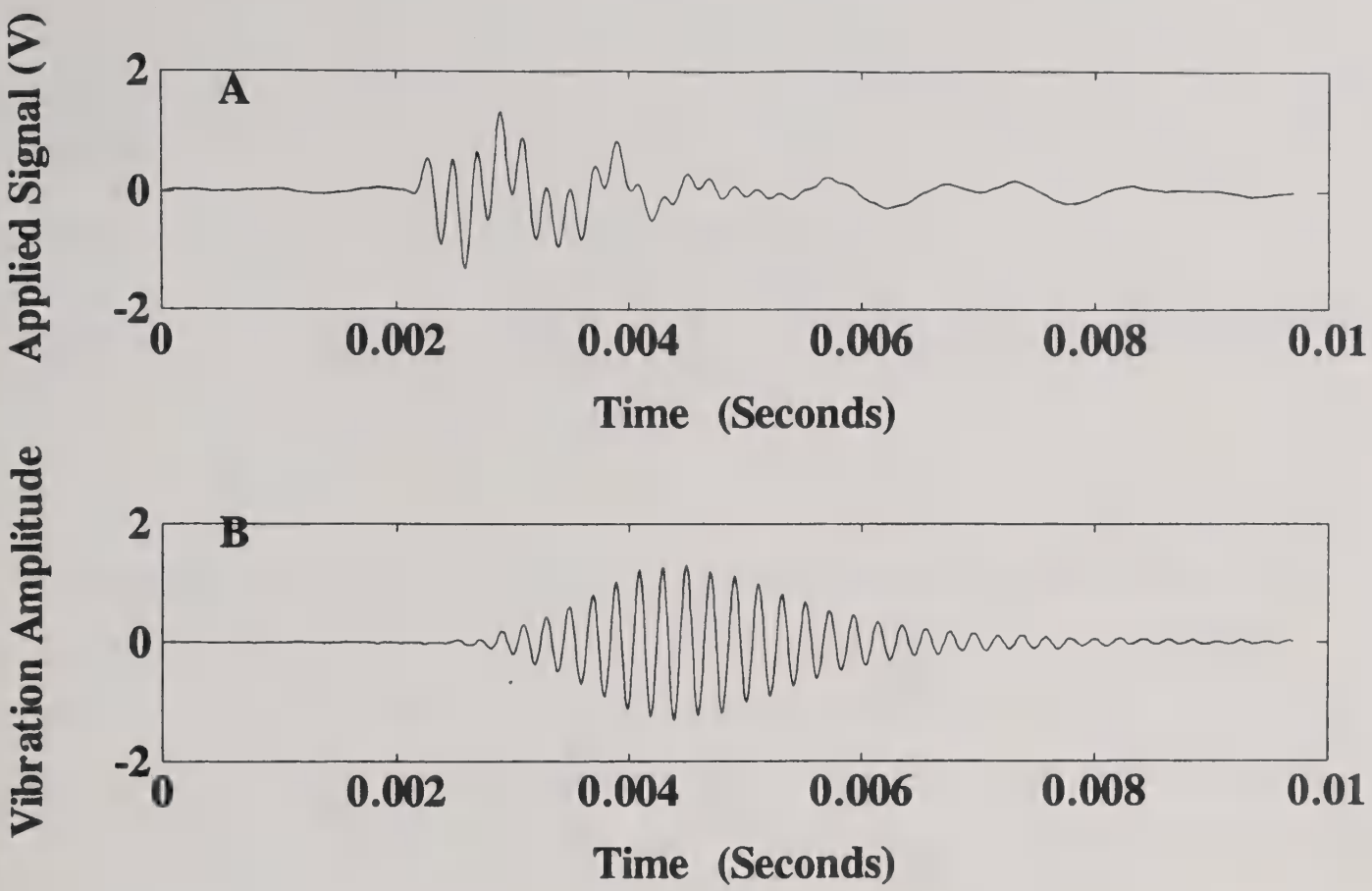

Figure 8

Fourth to the last applied vibration impulse (A) and corresponding detected vibration impulse (B).

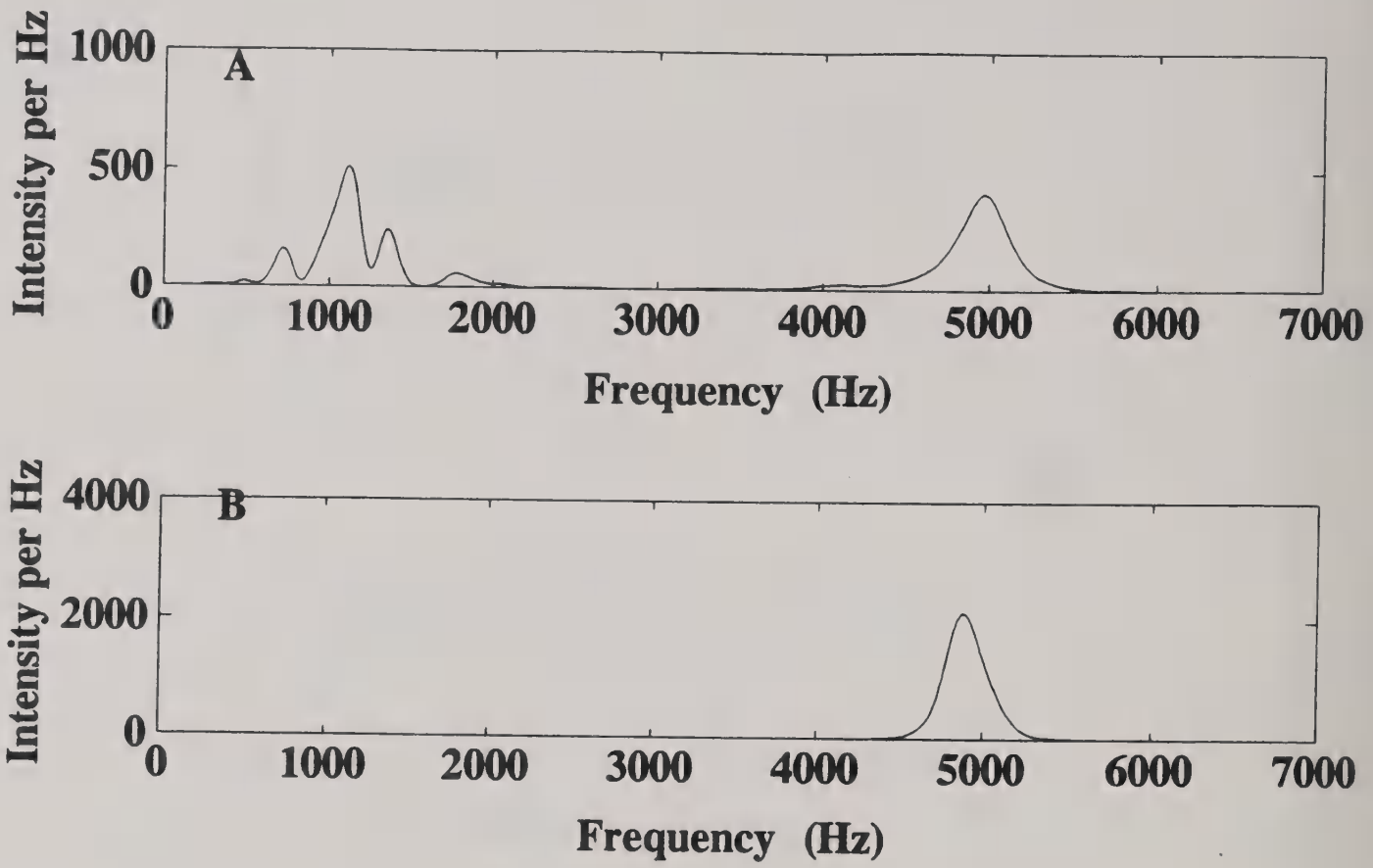

Figure 9

Frequency content of the impulses of figure 8. Part A is spectrum of the applied impulse while B is the spectrum of the corresponding detected substrate vibration impulse.

Biochemistry and Physiology of Overwintering in the Mature Larva of the Sunflower Stem Weevil, *Cylindrocopturus adspersus*.

ROBERT R. ROJAS, LAURENCE D. CHARLET* and ROGER A. LEOPOLD

Biosciences Research Laboratory and *Northern Crop Science Laboratory, USDA-ARS, Fargo, ND 58105, U.S.A.

The sunflower stem weevil (SSW), *Cylindrocopturus adspersus*, overwinters at the base of the sunflower stalk as a mature larva. Sunflower stalks from fields known to be infested with SSW larvae were collected in southeastern North Dakota in October 1991. Larvae from stalks kept outdoors accumulated a high whole-body concentration of trehalose (up to 69 µg/mg wet wt) at the expense of glycogen with the onset of winter followed by a partial reconversion of trehalose to glycogen with the onset of spring. Larvae from stalks acclimated to 0°C also accumulated a high level of trehalose (~69 µg/mg wet wt) with a concomitant decrease in glycogen. Those larvae from stalks acclimated to 20°C showed an initial sharp increase in whole-body trehalose that then stabilized but at a concentration well below that of larvae from stalks kept outdoors or acclimated to 0°C. This indicates that there exists in the larva an underlying developmental component to trehalose accumulation which is further enhanced by low temperature (0°C) exposure. The mean temperature of crystallization (T_c) of larvae exposed to outdoor conditions showed an abrupt drop from October (-25.0 ± 1.3°C) to November (-28.2 ± 0.6°C) with a minimum in February (-29.1 ± 0.3°C). The level of trehalose accumulated by the SSW larva is to our knowledge the highest reported in an overwintering insect.

Identification and Characterization of a Bacterial Toxin Active Against the Red Sunflower Seed Weevil.

W. Scott Grayburn

USDA, ARS, Northern Crop Science Laboratory, Fargo, ND 58105

Many insects have been controlled by use of a natural insecticide derived from the bacterium Bacillus thuringiensis (Bt). One of the main advantages of this approach is that different bacterial strains show toxicity against a limited group of insects. For this reason, different Bt strains have been used to control a wide variety of specific insect pests, usually in larval feeding stages. I previously screened various strains of Bt for toxicity against adult female and male red sunflower seed weevils (Smicronyx fulvus). Of 22 strains tested, one, a Bt israelensis strain, was found to be toxic against adult seed weevils (Grayburn, 1994).

Field applications of intact bacteria (Bt) have been successfully used to control a number of insects, and this approach may also work for seed weevil control. Ideally, the toxin from the Bt responsible for weevil mortality should be produced by sunflower, eliminating the need for field application.

Genes for delta endotoxins that are toxic to a limited range of target insects have been cloned from various Bt strains and the genes sequenced. Analysis of this sequence data revealed regions of DNA that were highly conserved among closely related strains (Carozzi et al., 1991). The Dip1A and Dip1B sequences are conserved among Bt israelensis strains. These oligonucleotides were used with the polymerase chain reaction (PCR) to amplify part of a delta endotoxin gene from the Bt israelensis strain that was toxic to adult seed weevils. This PCR product was cloned (plasmid pSBT1) and the ends were sequenced. As expected, a high degree of similarity was seen between this sequence and previously published DNA sequences for Bt israelensis delta endotoxin genes. Based on sequence data from pSBT1, oligonucleotide primers were synthesized to isolate an adjacent region of the endotoxin gene using 'inverse' PCR. This product will be cloned and partially sequenced. In the future, this new clone will be transferred to a bacterial expression plasmid, and the fusion protein will be tested for toxicity against adult seed weevils. If the fusion protein is found to be toxic, the gene fragment will be transferred to a plant expression plasmid and used for sunflower transformation experiments.

Selected References

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INSECT / PLANT / MICROBE INTERACTIONS: CURRENT RESEARCH AND FUTURE PLANS

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The banded sunflower moth *Cochylis hospes* was the focus of current studies and future projects on oviposition, natural sunflower resistance and induced sunflower resistance in the form of *Bacillus thuringiensis* toxin. In oviposition bioassays, aqueous extracts were found to be the most active. Banded sunflower moths responded in a dose response manner to odors released from aqueous extracts. A higher concentration of 0.33 gr / ml of bract tissue vs 0.16 gr / ml. of leaf tissue was required to obtain a peak in ovipositon. Aqueous extracts of hybrids were preferred to wild *Helianthus tuberosus*. Boiling the aqueous extract to test for stability and volatility of the odor that induces oviposition resulted in a loss of activity after 30 minutes. Activity was also lost if the extract was left standing for 24 hours at 4°C. Extracts prepared with non-aqueous solvents showed low activity possibly because of toxicity and possibly because of high volatility and loss in the preparation of the bioassay. To get information on putative oviposition stimulants or attractants that were soluble in non-aqueous solvents, 37 commercially available sunflower chemicals were bioassayed. Thirty four of the 37 tested negatively suggesting repellancy or inhibition by these compounds, at least, when they are presented to the moths in isolation from other sunflower chemicals. Electrophysiological recordings from sensory organs were discussed as a possible method of narrowing down the number of possible chemical odors that the banded sunflower moth responds to in its oviposition behavior. Another part of the sunflower plant that has been implicated as an important nutrient or attractant for some insects is pollen. Preliminary experiments indicated that the banded sunflower moth was repelled by pollen and further experiments to explore the interaction of banded sunflower moths with pollen are planned for the summer season. Laboratory experiments on natural sunflower resistance took the form of evaluating the incorporation of various sunflower parts into the diet used to maintain a laboratory colony. Florets, leaves, petals and sunflower seed were incorporated into the diet and mortality and changes in development with respect to control diets were observed. Only the oil fraction of the seed showed effects on development time and mortality. Further processing of the oily fraction and bioassay of the fractions are planned. Future projects include possible evaluation of suflower constructs with induced resistence in the form of Bt toxin produced by a gene incorporated into sunflower from the bacterium *B thuringiensis*.

DEVELOPMENT OF STRATEGIES FOR LABORATORY PRODUCTION OF RED SUNFLOWER SEED WEEVIL AND BANDED SUNFLOWER MOTH[1]

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Red sunflower seed weevil and banded sunflower moth adults are present only during the months of June, July and August in the northern plains. Thus, research utilizing adults developing from field larvae is limited to this period. The objective of this study was to develop culture techniques that provide adult insects from field collected larvae year-round.

MATERIALS AND METHODS

Sunflower heads naturally infested with either red sunflower seed weevil or banded sunflower moth larvae were collected from the field in late August to early September and brought to the laboratory. Last instar larvae dropping from the heads were collected, weighed or counted, and placed in rearing containers filled with sterilized soil. All tests were replicated four times.

Rearing containers were held for specific periods of time in a cold room at 4°C, then moved to a rearing room maintained at 26°C. All containers were watered twice weekly.

Red Sunflower Seed Weevil

In 1991, the effect of moisture, temperature, rearing container size, and larval population density on the development of red sunflower seed weevil larvae to the adult stage was tested. In 1992, media type and media weight were added to the list of variables tested in 1991. Data were collected on number of days from the time the larvae were put in the containers to first adult emergence and percentage of adults or parasites that emerged.

Moisture - In the moisture study, 3.75-L plastic containers were filled with sterilized soil and weighed. The total weight of the container and soil was 3000 grams. Approximately 300

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[1] U. S. Department of Agriculture, Agricultural Research Service, Northern Plains Area, is an equal opportunity/affirmative action employer and all agency services are available without discrimination. This paper reports the results of research only. Mention of a product does not constitute a recommendation for its use nor an endorsement of the product by the USDA.

larvae were put on the soil surface in each container. Containers were chilled at 4°C for 10, 16 or 22 weeks. Two levels of moisture were tested. Half the containers received 50 ml of distilled water twice weekly and and the other half received 30 ml.

Cold Treatment - Containers were cold-treated at -4°C for specific periods of time in 1991. They were then held at 4°C for 14 weeks before being transferred to the rearing room. In 1992, the containers were exposed to either -4°C or 0°C for specific periods of time, after which they were held at either 4°C, 26°C or 30°C. From 4°C, the containers were transferred to 26°C until adult emergence.

Container Size and Larval Population Density - Seven container sizes ranging from 185-ml vials to 18.75-L buckets were used in 1991. In 1992, only five sizes, also ranging from 185-ml vials to 18.75-L buckets, were used. Three different population densities designated as low, medium or high were tested in each container size. Actual number of larvae used per container size are shown in the table below. All containers were held at 4°C for 14 weeks, then moved to the rearing room.

Table 1. Larval population density (P) in each container size. (Except for C6 and C7 where larvae were counted, larvae were weighed so numbers are estimates based on the weight of 0.00395 g per larva.)

Container Size	P1=Low	P2=Medium	P3=High
C1=18.9 L	1000	2000	5000
C2=15 L	800	1500	3000
C3=11 L	500	1000	2000
C4=3.785 L	300	500	1000
C5=1 L	100	250	500
C6=.5 L	50	100	250
C7=185 ml	50	75	100

Media Type - Three different types of media were tested. One-liter containers filled with soil, Sunshine Mix® or Metro Mix®, and 100 weevil larvae were exposed to -4°C for 0, 2 or 4 weeks and then moved to 4°C. After 14 weeks, the containers were transferred to the rearing room.

Media Weight - One thousand, 2000, or 3000 grams of soil were put in 3-L containers. Two hundred weevil larvae were put on the soil surface in each container. They were held at 4°C for 14 weeks, then moved to the rearing room.

Banded Sunflower Moth

The effect of cold treatment on adult emergence was tested by exposing one-liter containers with soil and 50 larvae to either -4°C or 0°C for specific periods of time. The containers were held at -4°C, 4°C, then moved to 26°C until adult emergence. Rearing containers exposed to 0°C were transferred to either 4°C, 26°C, or 30°C. From 4°C, the containers were moved to 26°C after 18 weeks of cold treatment.

RESULTS AND DISCUSSION

Red Sunflower Seed Weevil

Moisture - Results from the moisture tests indicated that the number of days to first adult emergence depended on the length of time the larvae were held in cold storage. In 1991 (Fig. 1), except for containers that were chilled for 16 weeks and watered twice weekly with 50 ml of distilled water, the number of weevil adults that emerged from the moisture treatment did not differ significantly. In 1992 (Fig. 2), the amount of moisture added to the containers did not have a significant effect on either weevil or parasite emergence.

Cold Treatment - In 1991 (Fig. 3), the number of days the containers were held at -4°C did not have a significant effect on the date of the first adult emergence or the percent of weevil or parasites that emerged. In 1992, transfer of containers from both -4°C (Fig. 4) and 0°C (Fig. 5) to either 26°C or 30°C significantly lowered weevil and parasite emergence compared to those that were held at 4°C then moved to 26°C. However, duration of larval diapause to adult emergence was shortened. In general, exposure of rearing containers to temperatures below 4°C lowered percent adult and parasite emergence.

Container Size and Larval Population Density - Results in 1991 (Fig. 6) showed that the lower the population density in a particular container, the higher the percent adult emergence. Highest percent adult emergence was obtained from 185-ml vials. However, in 1992 (Fig. 7), no correlation was observed between larval population density and percent emergence. Eleven-liter containers yielded the highest percent adult emergence.

Media Type - The type of media had no effect on the number of days to first adult emergence but there was a significant effect on percentage of both weevil and parasite emergence. Soil was the best media for the production of adults (Fig. 8).

Media Weight - The amount of media (soil) did not affect the number of days to first adult emergence or the percent emergence of weevil adults and parasites (Fig. 9).

Banded Sunflower Moth

Exposure of rearing containers to -4°C did not result in significant differences in the number of days to first adult emergence or the number of moths or parasites that emerged (Fig. 10). However, direct transfer of rearing containers from 0°C to either 26°C or 30°C significantly shortened larval diapause and development to the adult stage while longer exposure to 0°C had the opposite effect (Fig. 11). The percent emergence of moths and parasites was significantly reduced by prolonged exposure to 0°C or direct transfer of containers to 26°C and 30°C.

CONCLUSIONS

Red Sunflower Seed Weevil

Thirty milliliters of distilled water added twice weekly to 3000 g of soil gave the highest percentage adult emergence. The number of days to first adult emergence was dependent on the length of time the rearing containers were held in cold storage. Exposure of containers to 4°C for 10, 16 or 22 weeks did not result in a significant difference in adult emergence.

Chilling the containers at 4°C gave the highest percent weevil emergence. Exposure of containers to temperatures below 4°C had no impact on the number of days to first adult emergence.

Red sunflower seed weevil and parasite emergence varied widely among the different sizes of rearing containers with different larval population. No consistent effect was seen.

Soil was the best media for rearing red sunflower seed weevil larvae to the adult stage. Using different soil weight did not have an effect on the date of first adult emergence and percent weevil or parasite emergence.

The best conditions for red sunflower seed weevil emergence were: 11-L containers filled with sterilized soil; 500 larvae per container; containers chilled for 98 days at 4°C and then held at 26°C; and watered twice weekly.

Banded Sunflower Moth

Exposure of rearing containers to -4°C did not affect the number of moths or parasites that emerged or the number of days to first adult emergence. Containers held at 4°C for 18 weeks gave the highest percent moth emergence. The shortest period of time for larval diapause and development to adult was obtained when containers were held at 0°C for four weeks, then moved to 30°C.

Results showed that parasitism of banded sunflower moth larvae was high. The number of parasites emerging in most of the test containers was greater than the number of moths that emerged.

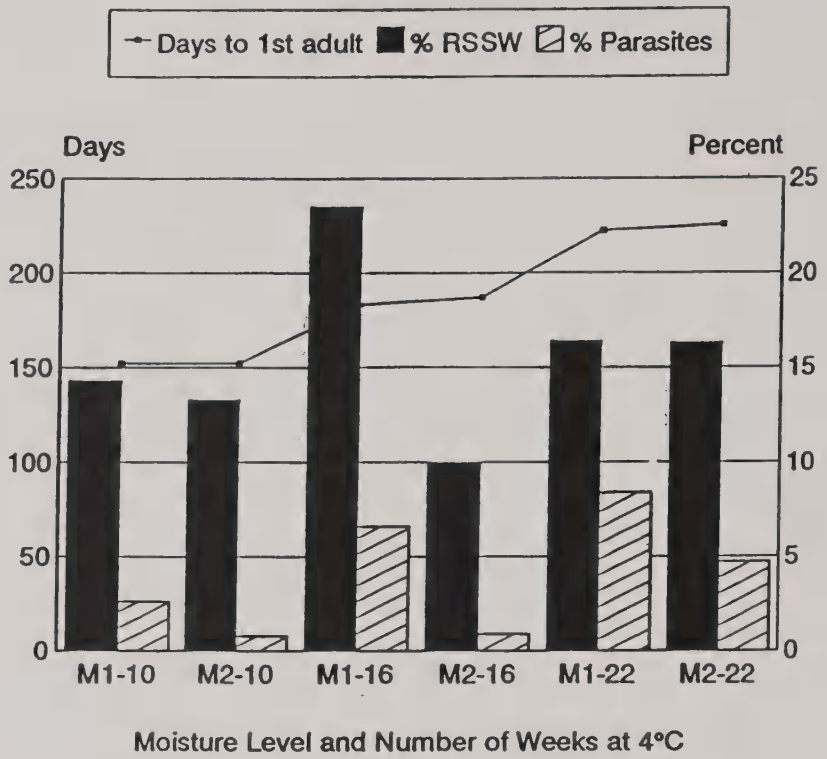

Fig. 1. Moisture effect on red sunflower seed weevil and parasite emergence, 1991. (M1, M2 = 50 or 30 ml distilled water added twice weekly to each container).

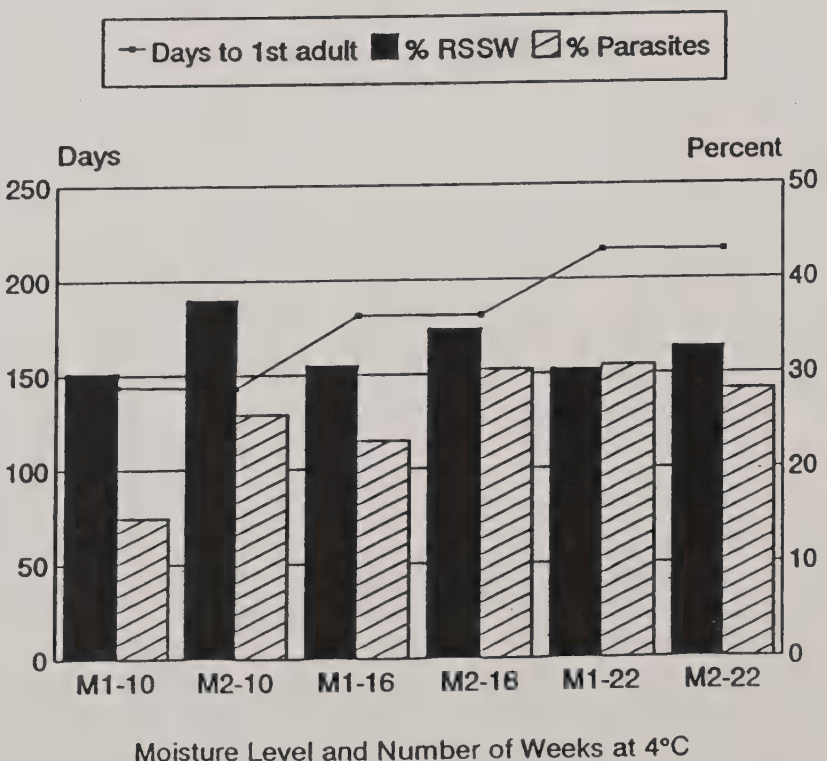

Fig. 2. Moisture effect on red sunflower seed weevil and parasite emergence, 1992. (M1, M2 = 50 or 30 ml distilled water added twice weekly to each container).

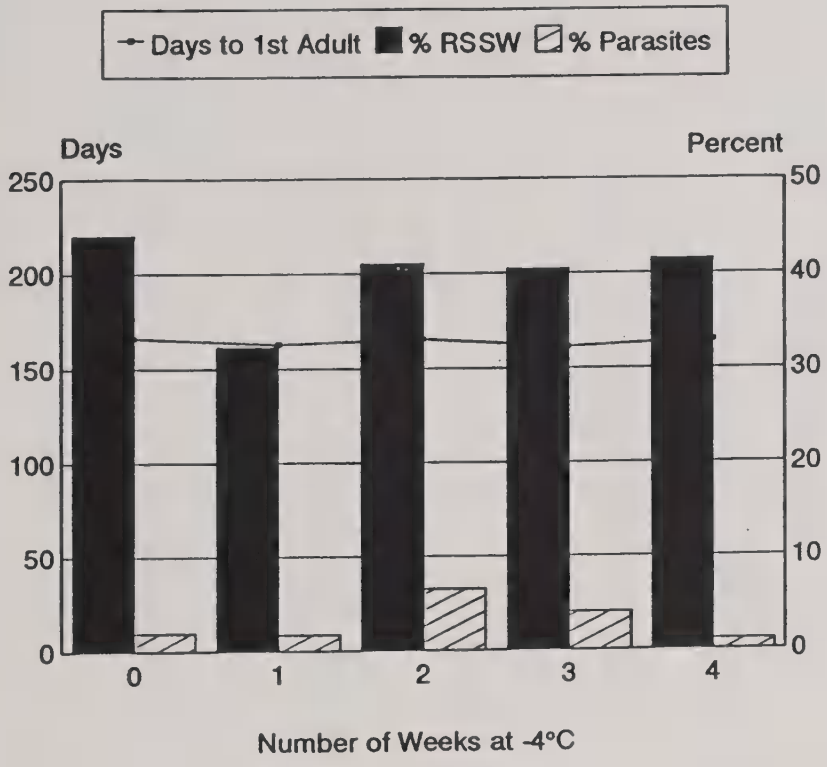

Fig. 3. Effect of exposure to -4°C on red sunflower seed weevil and parasite emergence, 1991.

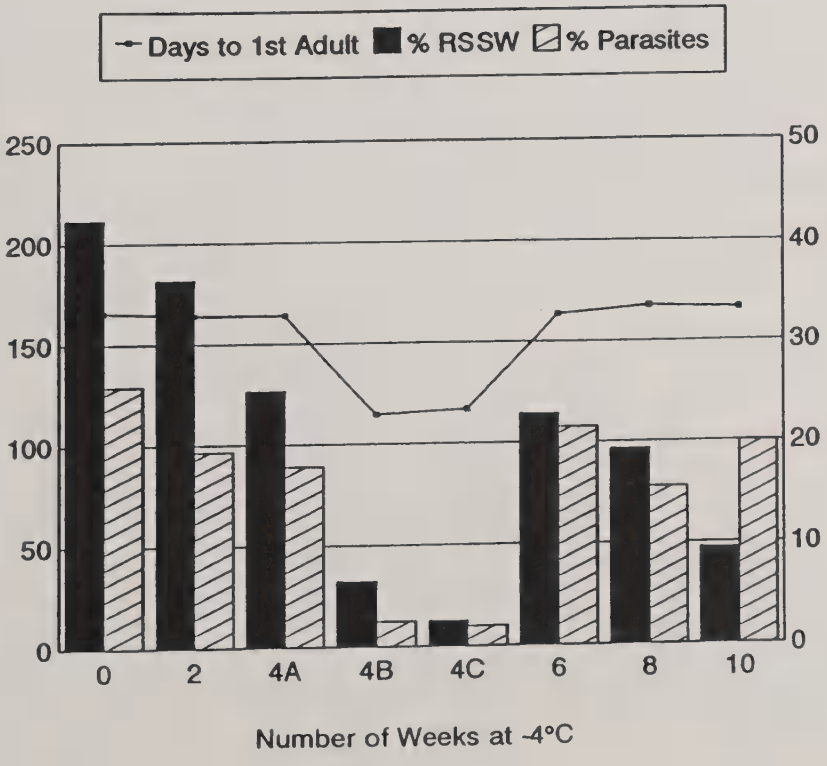

Fig. 4. Effect of exposure to -4°C on red sunflower seed weevil and parasite emergence, 1992. (4A, 4B, 4C = moved to 4°C, 26°C or 30°C after 4 weeks, respectively).

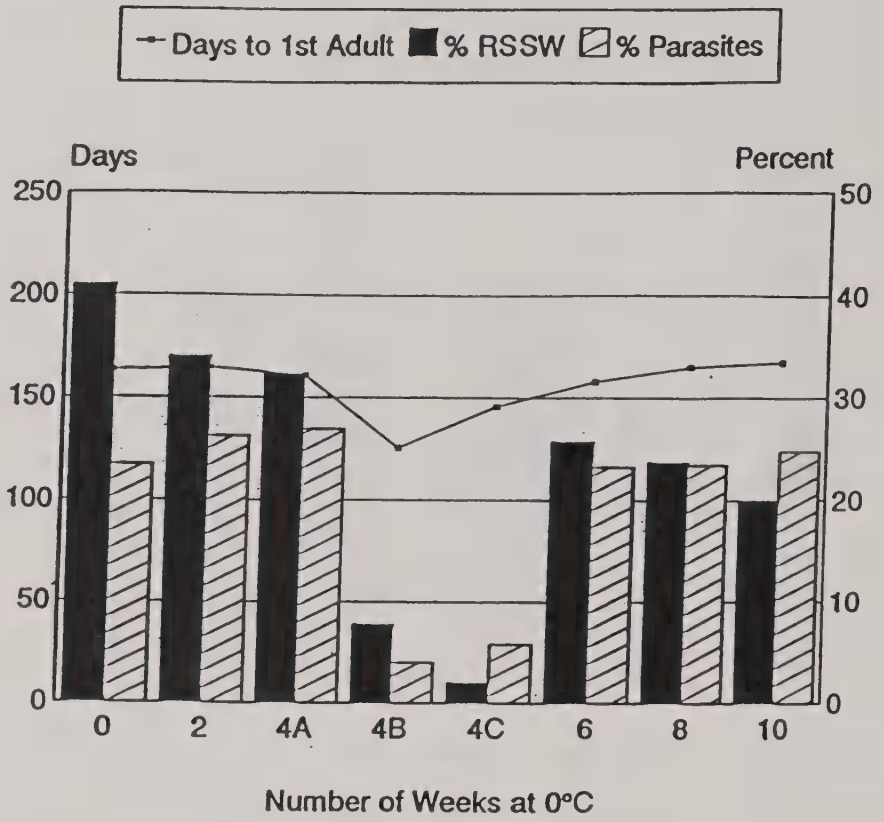

Fig. 5. Effect of exposure to 0°C on red sunflower seed weevil and parasite emergence, 1992. (4A, 4B, 4C = moved to 4°C, 26°C or 30°C after 4 weeks, respectively).

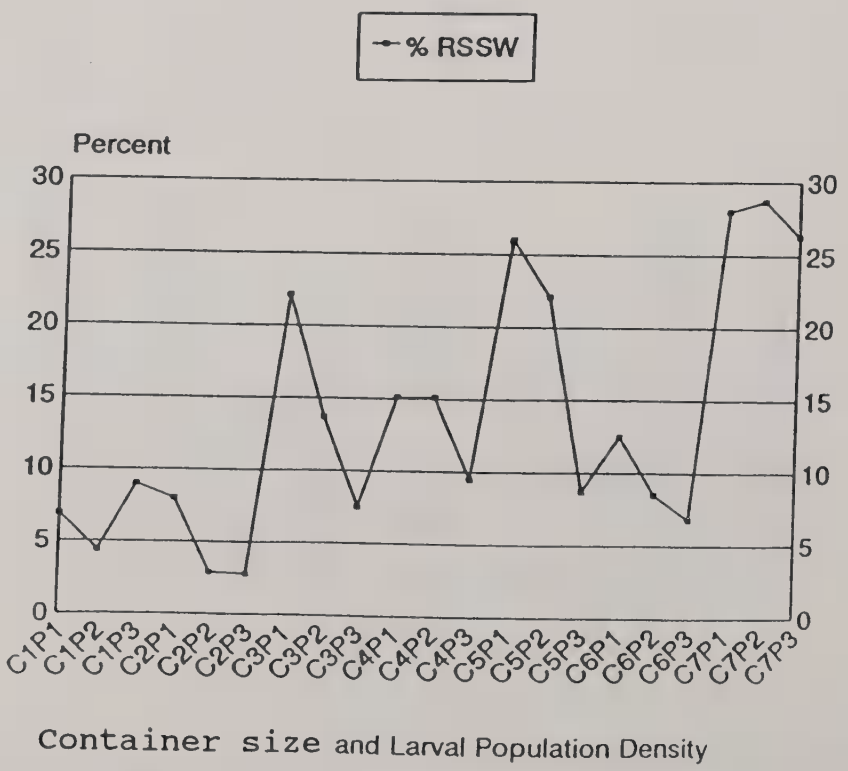

Fig. 6. Container size and larval population density effect on red sunflower seed weevil emergence, 1991. (C1=18.9L; C2=15L; C3=11L; C4=3.785L; C5=1L; C6=.5L; C7=185ML; P1, P2, P3 = low, medium or high larval population density, respectively).

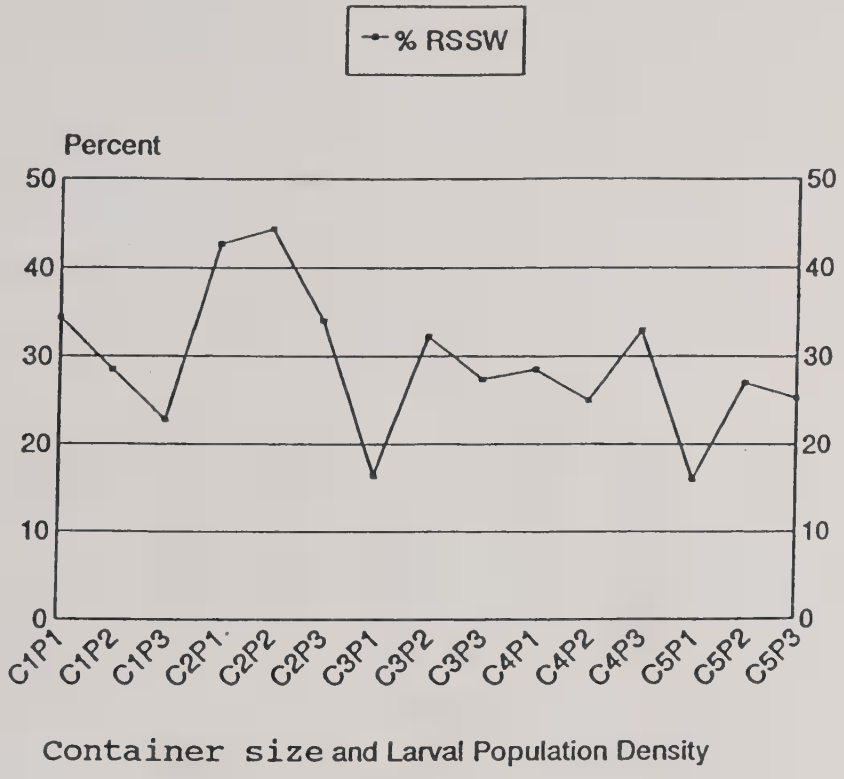

Fig. 7. Container size and larval population density effect on red sunflower seed weevil emergence, 1992. (C1=18.9L; C2=11L; C3=3.785L; C4=1L; C5=185 ML; P1, P2, P3 = low, medium or high larval population density, respectively).

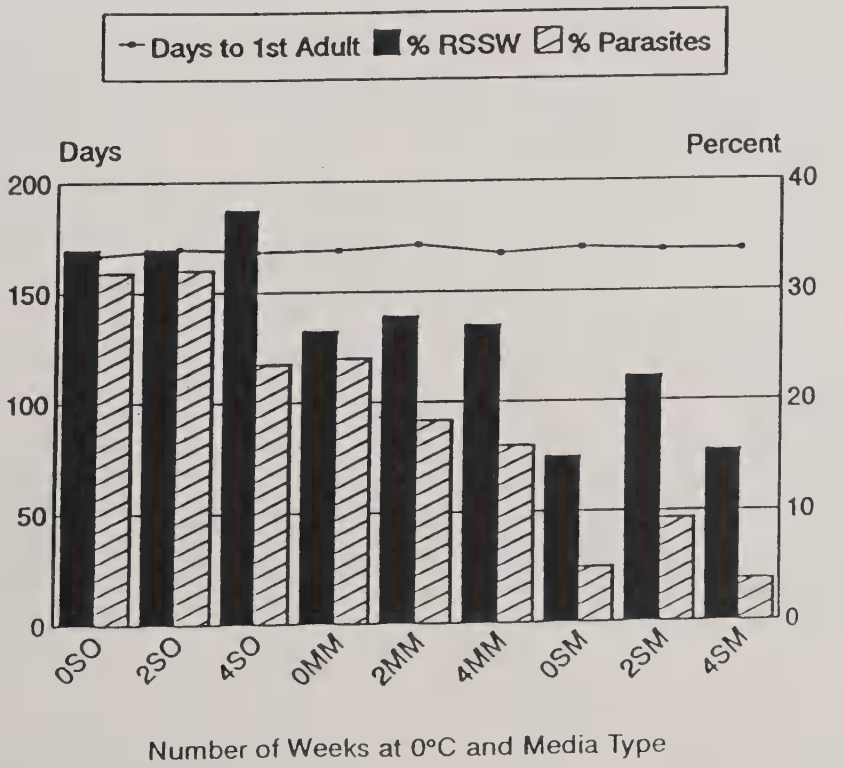

Fig. 8. Red sunflower seed weevil and parasite emergence on different media, 1992. (So=Soil; MM=Metro Mix®; SM=Sunshine Mix®).

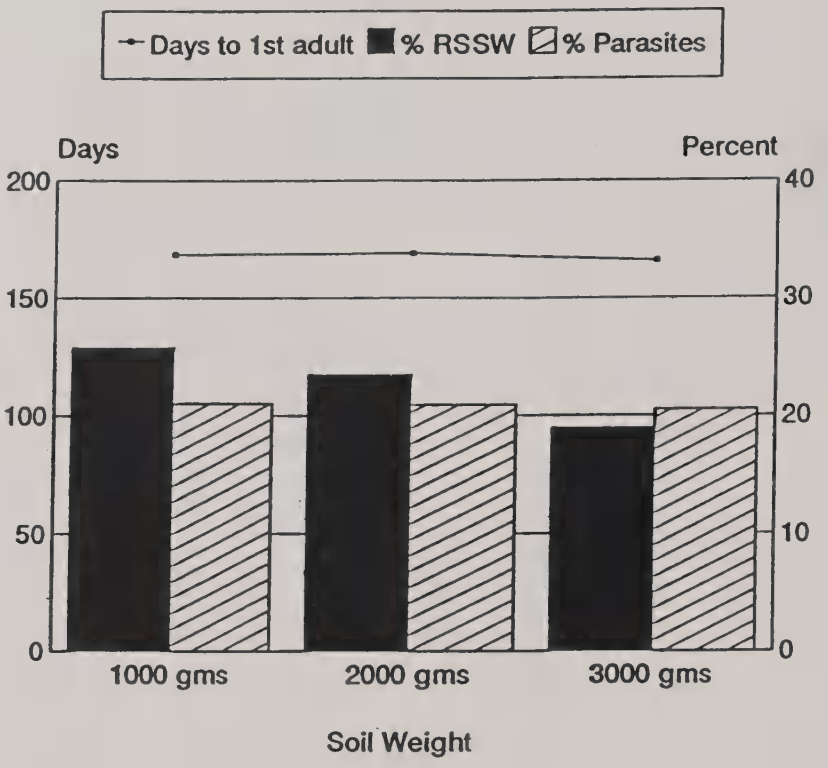

Fig. 9. Effect of media weight on red sunflower seed weevil and parasite emergence, 1992.

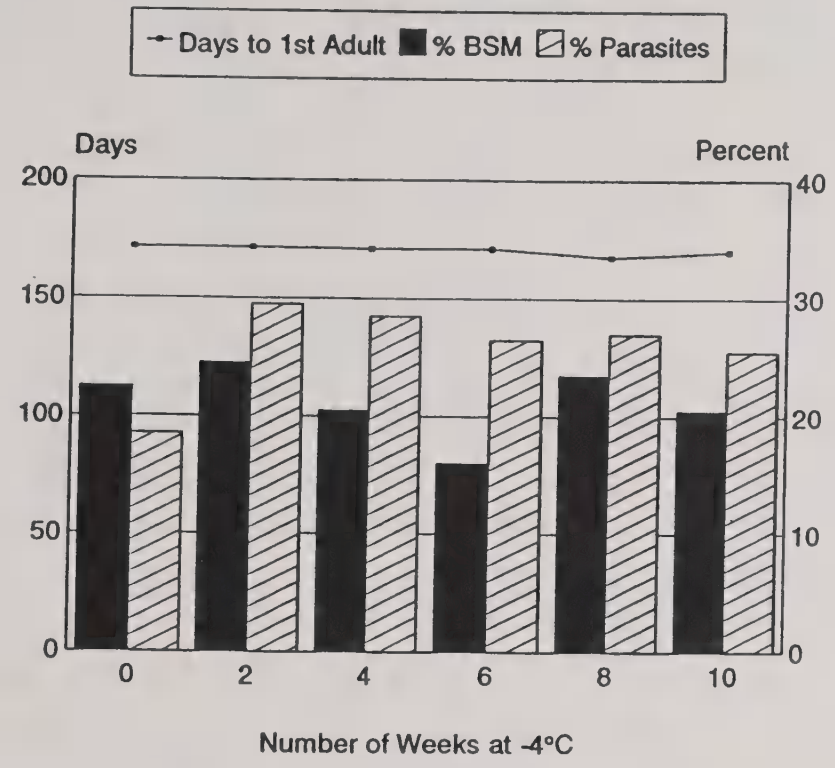

Fig. 10. Effect of exposure to -4°C on banded sunflower moth and parasite emergence, 1992.

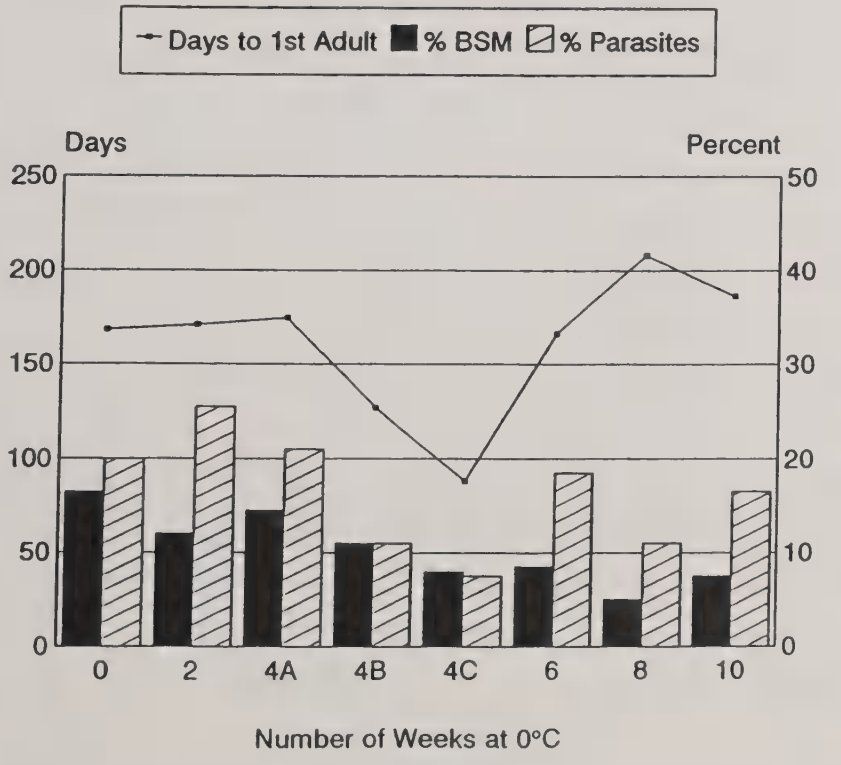

Fig. 11. Effect of exposure to 0°C on banded sunflower moth and parasite emergence, 1992. (4A, 4B, 4C = moved to 4°C, 26°C or 30°C after 4 weeks, respectively).

Evaluation of the National Plant Germplasm System Cultivated and Wild-type Sunflower Germplasm for Resistance to Sunflower Moth

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The sunflower moth, *Homoeosoma electellum* (Hulst), is a very serious pest of commercial sunflower in the United States. One environmentally-friendly method for controlling this pest is growing cultivars resistant to sunflower moth feeding. Currently, host plant resistance to this pest has not been extensively incorporated into commercial cultivated sunflowers. This paper describes techniques for evaluating the cultivated and the wild-type sunflowers in the National Plant Germplasm System's collection of sunflower germplasm at Ames, IA for resistance to the sunflower moth.

Evaluation of cultivated types. Each accession to be tested is planted in a 25 ft row. When the plants reach the R5.2 stage, 2 or 3 egg pads (each with ca. 30 eggs) are pinned to 5 test heads. One week after infestation, 5 check heads are sprayed with Asana. One day after spraying, muslin bags are placed on the 5 egg infested heads and on the 5 sprayed check heads. The heads are left in the field and are harvested at maturity. The diameters of the heads are measured before hand threshing and achene processing (to remove the inferior achenes). The high quality achenes are weighed and counted. Calculations made are area of head (cm^2), number of high quality achenes/cm^2, and weight of high quality achenes/cm^2 for both infested heads and check heads. A comparison of the damaged heads to the check heads is made using the following: number (or weight) of high quality achenes on infested heads/ number (or weight) of high quality achenes on check heads X 100 = % of control.

Evaluation of wild-types. Since wild sunflowers are multiple-headed, they must be handled differently. Accessions to be tested are started in the greenhouse and later transplanted to the field. Each accession is planted into 2 plots of 20 plants each and covered with cages measuring 20 ft long x 10 ft high x 10 ft wide. Both cages receive a nuc box of honey bees (ca. 4000 bees with a queen) to aid in pollination. Plants in one of the cages will receive 100 sunflower moth pupae when they begin to flower. When the plants are mature, they are hand harvested and the seed are processed to remove the inferior achenes. Total seed weights are measured for each cage. Comparisons are made by dividing the weight of high quality achenes in the sunflower moth infested cage by the weight of high quality achenes obtained in the check cage.

Screening Sunflower Germplasm for Resistance to the Seed Weevil

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Screening sunflower germplasm (*Helianthus annuus* L.) for resistance to the red sunflower seed weevil (*Smicronyx fulvus* LeConte) and determination of the mechanisms of resistance were done in 1993.

In 1993, 24 lines were planted at Prosper, ND in a RCBD with 4 replications. Hybrid 894 was used as a standard check. A lepidopteran specific insecticide, B.T., was applied from sunflower stage V2 to stage R5.3 to control a banded sunflower moth infestation. When sunflower reached R5.5-5.6, 2 heads of each variety in each block were artificially infested with 30 adults and enclosed with Delnet (R) pollination bags to prevent the weevils from escaping. Three days later, these artificially infested heads were uncovered and sprayed with malathion to kill the weevils. In late August of 1993, the infested heads were covered with larvae collecting bags to capture emerging, mature larvae. In the middle of October, the artificially infested sunflower heads and two control plants per row in each plot were harvested. The mean number of larvae per sunflower head was determined and analyzed by SAS.

Of the germplasm lines tested, differences in number of larvae collected were found (Fig. 1). The number of larvae emerging ranged from 61 to 18 per head. These lines will be tested again using both artificial and natural infestation.

To test for adult nonpreference, 6 germplasm lines (Fig.2.) were selected based on the 1992 field results and tested a greenhouse using a RCB design. At stage R5.5, the plants were arranged in a circle in a 1.8 X 1.8 X 1.8 m cage and about 360 newly emerged adult weevils were released into the center of each cage. One plant of each line was placed in each cage, and 3 cages were used. After 10 days, the adults on each plant were counted and removed. Among the 6 germplasm lines tested, 894 was most attractive (Fig.2). Thus, by both methods of resistance

evaluation, free choice test and artificial infestation test, hybrid 894 was susceptible. Line 170411 was highly attractive (Fig. 2), however, this was unexpected because it had a low infestation when artificially infested (Fig. 1). These experiments will continue in 1994 and 1995.

Fig. 1. Mean no. of larvae per sunflower head on selected sunflower germplasm lines artificially infested in a no-choice test (P = 0.094)

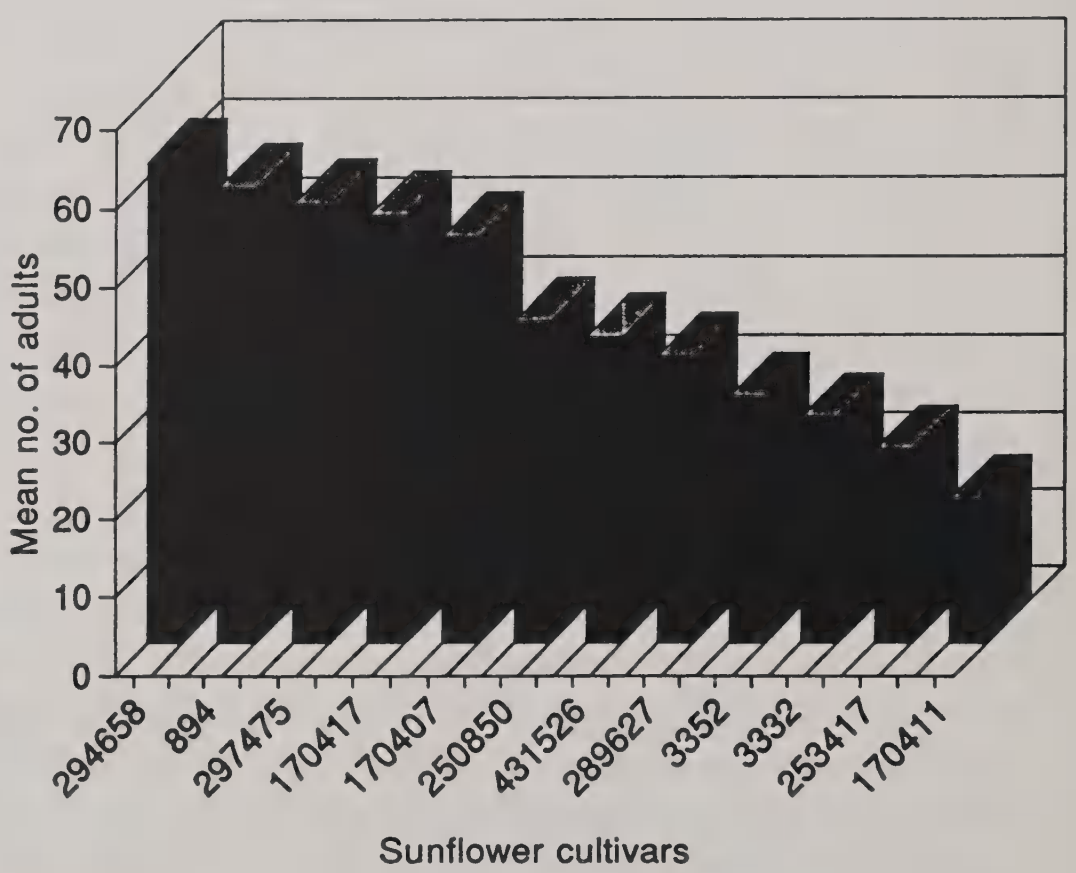
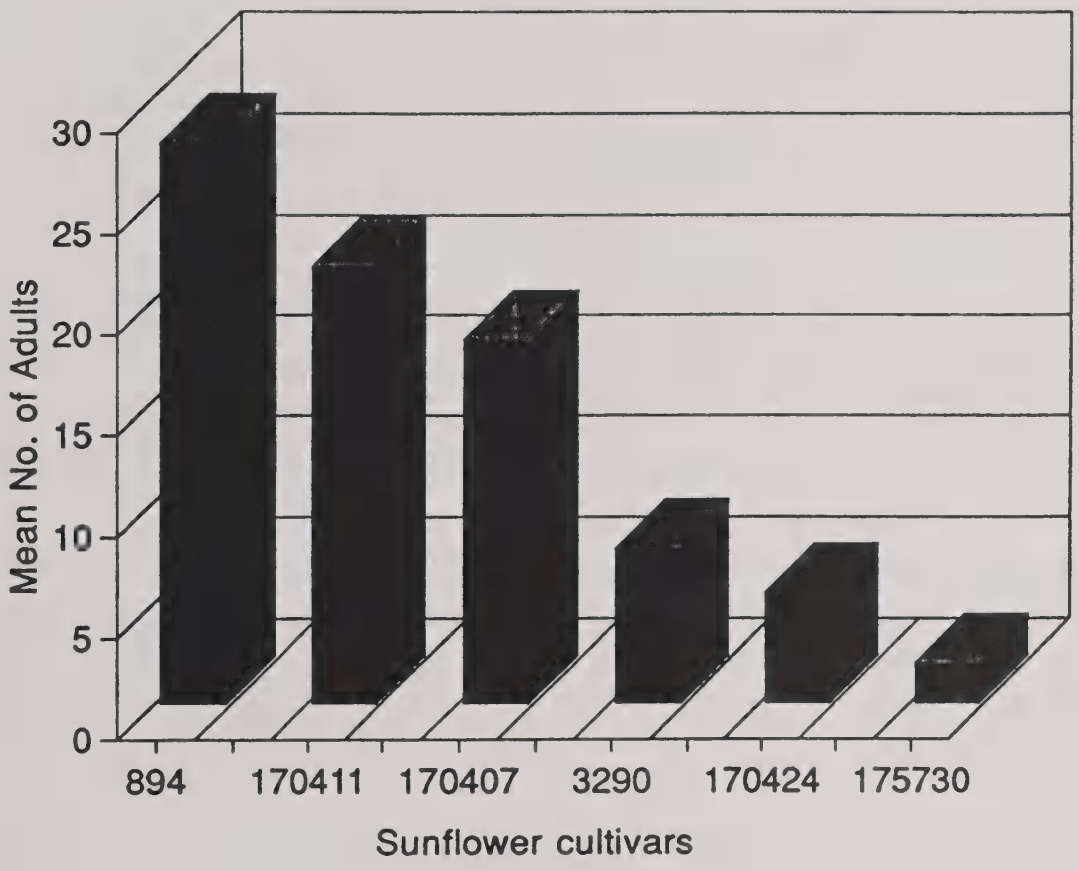


Fig. 2. Mean no. of rssw adults on different germplasm lines tested in a free choice test in greenhouse. Spring, 1993.



Trap cropping to manage the red sunflower seed weevil

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Intercropping is the planting of more than one crop in a field with the goal of improving yield by the interaction of the different crops. A trap crop is a special type of intercrop where the intercrop attracts the pest away from the main crop (Vandermeer 1989) and is designed to increase yield by lowering pest damage. Trap crops may be intercropped with the same or a different species. Risch et al. (1983) found that a low pest population will maximize yield when competition among the intercrops is minimal.

Trap crops may be of one of two designs. In the first type termed edge trapping, a trap border is planted around the entire field that is to be protected. In the second type, blocks or entire fields are planted to the trap crop and the fields being protected are not surrounded by the trap crop. In the second type of design it is important to know the location of the infestation source so that the trap field can be planted to intercept the pests as they are moving into the fields (Hokkanen 1991). In all cases, the area devoted to the trap crop is a small fraction of the field area being protected.

Trap cropping works because pests have distinct preferences for certain plant species and stages (Hokkanen 1991). The motivation for trap cropping is difficult or costly pest protection. Of the studies reporting economic benefits for trap cropping, a 10-30% overall increase in net profit was seen (Hokkanen 1991). Profitability is increased if the trap and the main crop are the same species, then both can be harvested together and the trap rows contribute to yield. To be effective a trap crop must be more attractive than the main crop for a least some critical period (Hokkanen 1991).

The red sunflower seed weevil, *Smicronyx fulvus* LeConte, causes severe economic loss to sunflower grown in the tri-state region of North and South Dakota and Minnesota (Oseto & Burr 1990). Insecticide applications are designed to minimize oviposition by the red sunflower seed weevil. The red sunflower seed weevil prefers sunflower that are in the blooming stage and that have seeds of intermediate maturity for oviposition (Brewer 1991) and they leave plants no longer producing pollen (Oseto & Braness 1979). Because sunflower is rotated each season, overwintering weevils emerging from the soil must migrate to new fields. If as they locate a field, a portion of the field is blooming, it will be preferred by the migrating seed weevils and may act as a trap.

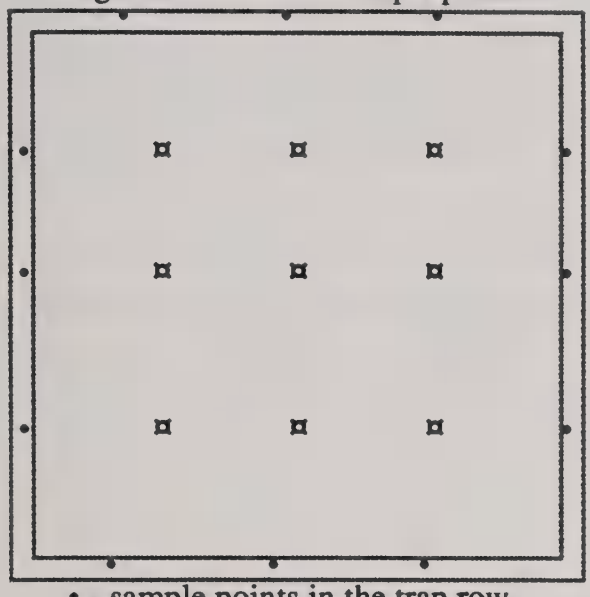
The objectives of this study were to determine if an edge of early blooming sunflower could serve as a trap crop for the red sunflower seed weevil. Specific objectives were to determine how effective the trap was in concentrating adult red sunflower seed weevils and how effective the trap fields were in preventing economic loss due to rssw infestation.

Methods

All testing was done on farms in North and South Dakota and Minnesota on commercial, oilseed production fields. Farmer cooperators planted one field as a trap and a second field as a check. Each cooperator planted trap rows around the outside of the trap field to an early maturing hybrid. The planting date was adjusted so the trap rows bloomed in late July to early August and began blooming 7 to 10 days earlier than the field interior. The trap was 16 rows wide. The

interior of the trap field and the check field were planted to a later season hybrid. When most plants in the trap rows reached stage R5.4 (40% of the head was shedding pollen), the cooperators counted the number of adult weevils per plant in the trap rows and in the field interior. Each field was systematically sampled at 21 points (Fig. 1). At each sample point, the 5 most mature plants were sampled. The trap rows were treated with insecticide shortly before the field interior began to bloom to control the adult weevils before they dispersed to the field interior. If weevil populations in either the trap field interior or the conventional field exceeded the economic threshold (Oseto & Braness 1980), they were treated with an insecticide.

Fig 1. Adult weevil sample points.



• - sample points in the trap row
¤ - sample points in field interior

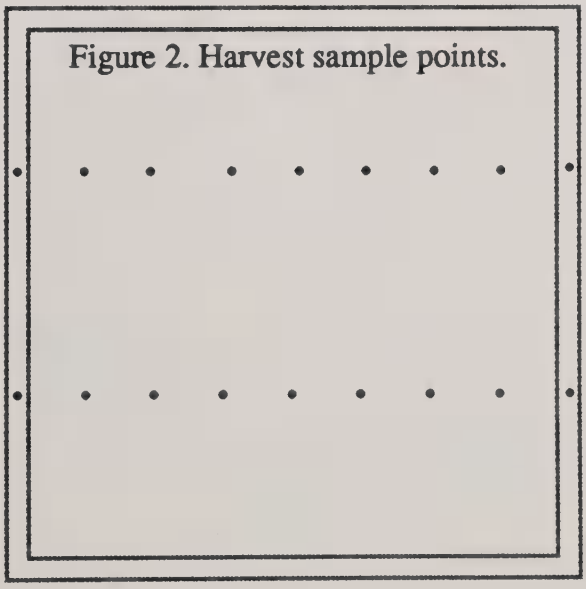
At plant maturity, samples were taken from 9 equally spaced sampling sites laid out as a row across each field. Two sampling rows, equidistant from the field ends, were sampled for a total of 18 sites per field (Fig. 2). At each sampling site, 5.3 m of row was harvested. Harvested plants were hand threshed and evaluated for yield and percentage of infested seed.

Based on the harvest data, the dollar loss attributable to red sunflower seed weevil infestation was determined. Preliminary testing indicated that infested seeds weighed 0.028g (W_I) and uninfested seeds weighed 0.038g (W_U). The percent infested seeds was designated, %I; the percent uninfested seeds was designated, %U. For each field, the following calculations were made for prices of 0.18, 0.26, and 0.35 dollars per kg of seed.

(1) Infested seed / ha (INF) = yield/(W_I+ (%I x W_U)/%I)
(2) Yield loss / ha (YLD) = (INF x W_U) - (INF x W_I)
(3) Dollar loss / ha = (YLD x price) + (YLD x %I x 11.06 x price x 0.02)

Equation 3 takes into account loss due to weight reduction and loss due to reductions in oil percentage in infested seeds. Analysis of variance was used to compare the dollar loss in conventional vs. trap fields. Computations were made using direct yield loss alone (weight and oil reductions) and yield loss plus insecticide cost.

Figure 2. Harvest sample points.



Results

Over a three year period, 19 trials were established but data was collected from only five farms. In some cases the cooperators treated the entire trap field with an insecticide because of concerns about high weevil populations or weather conditions did not permit proper spray timing. All trials in 1993 failed either because of low weevil populations or flooding. Field size on the five farms for which data was obtained ranged from 22 to 60 ha. Calculations of the area of each trap field devoted to the trap edge ranged from 5.9 to 9.9%, with a mean of 7%.

Calculations were made assuming rectangular shaped fields, however, the fields had irregular borders because of sloughs and other obstructions. Thus, the actual field area devoted to trap edges is slightly greater than the calculated mean.

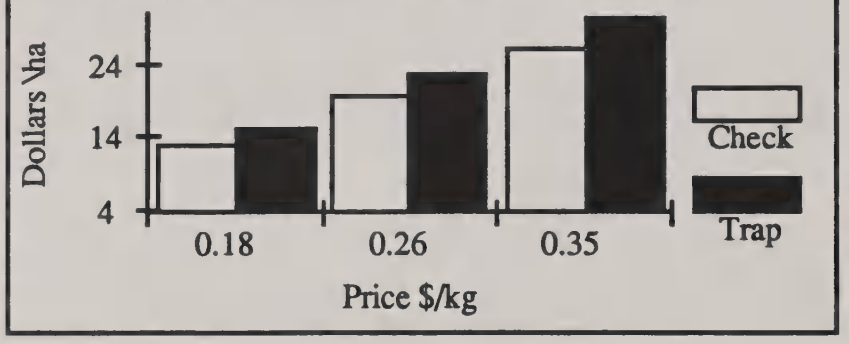
Significantly more red sunflower seed weevils were found on plants in the trap border than in the field interiors. A mean of 37 weevils per plant were on the plants in the trap border and 10.8 per interior plant. Although the difference was not significant, the mean seed infestation in the trap fields was higher than infestations in the check fields (Table 1).

Table 1. Mean percentage of seed infested by larval red sunflower seed weevils in five pairs of sunflower fields.

	Farm					
Field Type	DD91	JF91	DR91	DR92	DI02	Mean
Trap	10.3	28.9	08.4	08.4	04.9	12.2
Check	31.8	12.8	02.5	02.8	02.8	10.5

Although the difference was not significant, dollar loss directly attributable to rssw infestation was slightly higher in trap fields than in conventional fields (Fig. 3). However, when insecticide purchase and application costs were considered, the control fields had a greater dollar loss than did the trap fields (Fig. 4). But, the difference was not significant.

Figure 3. Dollar loss attributable to red sunflower seed weevil damage in check and trap fields.



Discussion

Red sunflower seed weevil damage in control fields was slightly less than that in the trap fields and this resulted in a higher dollar loss due to yield and oil reductions in the trap fields than in the check fields. However, the loss was slight and inconsequential when the cost of controlling the red sunflower seed weevil was considered. By treating an entire field when sunflower is priced at $0.18 / kg a grower would gain a yield increase valued at $2.09 / ha. However, to gain thet $2.09 / ha the grower had to treat the entire field with insecticide. Insecticide cost for a conventional field is $18.53 / ha and only $1.85 / ha for a trap field. Thus, the $2.09 gain cost an additional $16.68. When sunflower is of a higher value ($0.35 / kg), the gain for whole field spraying was $4.18. Still much less than the cost of whole field spraying.

Although trap cropping to control the red sunflower seed weevil is economical compared to whole field spraying, several considerations are important. First, the procedure is recommended for oil-type sunflower only. Marketing of edible seed sunflower allows a maximum seed infestation of approximately 3% to avoid a dockage penalty and infestations higher than 5-6% are rejected. Trap cropping did not lower seed infestations sufficiently, thus, it is not recommended to control red sunflower seed weevil infestations in edible seed sunflower.

Economic infestations may develop in the interiors of trap fields after the trap edges have been treated. This may occur because some weevils are in the interior when the edges are treated and are not controlled and some new weevils will migrate to the fields following edge treatment.

Thus, field interiors should be monitored to detect the possible development of an economic red sunflower seed weevil population.

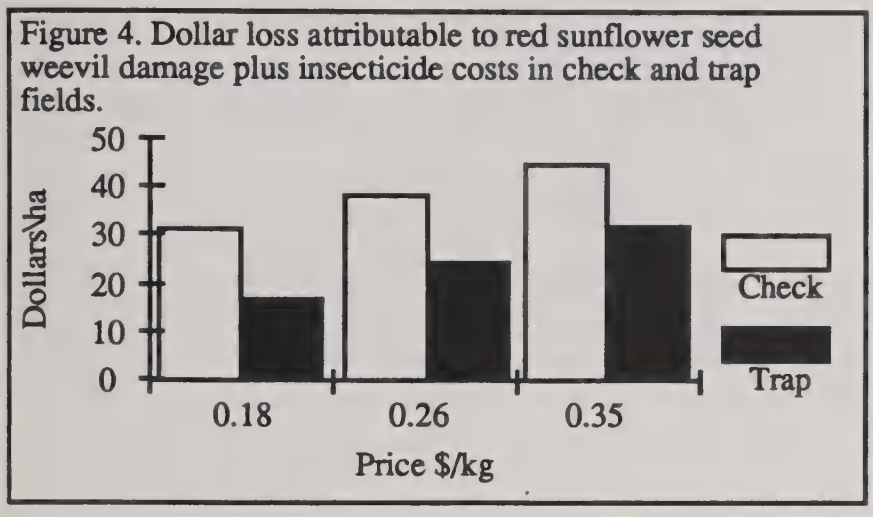


Figure 4. Dollar loss attributable to red sunflower seed weevil damage plus insecticide costs in check and trap fields.

However, for oil-type sunflower, trap cropping to manage the red sunflower seed weevil is recommended. The traps were effective in concentrating adult weevils along the field edges where they were easily controlled. Despite a slightly higher mean seed infestation and dollar loss due to infestation in trap fields, trap fields are economically advantageous because the cost of treating trap fields is approximately about 10% of that of whole field treatment.

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North Central Regional Plant Introduction Station's Sunflower Collection

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The National Plant Germplasm System's (NPGS) sunflower collection is maintained at the North Central Regional Plant Introduction Station (NCRPIS), Ames, IA. The 3608 sunflower accessions represent 8.5% of the total number of accessions maintained at the NCRPIS. The collection consists of 1445 cultivated *Helianthus annuus* accessions, 1406 wild annual *Helianthus* species, and 757 accessions of perennial *Helianthus* species.

The NCRPIS sunflower collection is the largest and most genetically diverse *ex situ* sunflower collection in the world and is vital to the conservation of *Helianthus* germplasm. Our acquisition activities adhere to the NPGS policies and are based on advice from the Sunflower Crop Advisory Committee. New accessions are acquired to fill 'gaps' in the total genetic diversity of the collection. These 'gaps' may be identified through species representation in the collection, through passport data including origin and habitat, and/ or through molecular techniques including isozyme, RFLPs, and RAPDs.

Acquisition activities have lead to a steady increase in the total number of sunflower accessions (Table 1). In 1993, 24 new *Helianthus* accessions were received at the NCRPIS. The average rate of growth from 1989-1993 was approximately three percent per year.

Our controlled-pollination and maintenance practices are designed to preserve the genetic integrity of the accessions. Hand-pollinated field regenerations are conducted for cultivated *Helianthus* accessions whereas wild annual and perennial *Helianthus* species are caged and pollinated using honey bees. Controlled pollinations of cultivated sunflower accessions are achieved relatively easily and, once accessions are increased, the seed stores relatively well. Maintaining and regenerating the wild sunflower accessions at the NCRPIS is difficult and expensive. Cages, bees, and the labor requirements for the maintenance of the perennial field nursery are all costly.

From 1989-1993, 15542 *Helianthus* packets were distributed to both domestic and foreign researchers (Table 2). During the past year, 1236 accessions, or 34.5% of the collection, were distributed for research purposes. The distribution of the collection is reflected in the availability of accessions. At present, 1268 accessions [775 (54%) cultivated *H. annuus* accessions and 493 (23%) wild *Helianthus* accessions] or 35% of the total collection are available for

distribution to the international scientific community. With the current facility and personnel resources, it will require about nine growing seasons to make all accessions available. Multiple regeneration sites are necessary to maintain and increase accessions not adapted to the environmental conditions at the NCRPIS.

Characterization data are compiled for relatively highly heritable floral, agronomic, and achene traits. Characterization and evaluation data are used to identify appropriate accessions for germplasm requests. All passport, characterization, and evaluation information for the Sunflower collection is maintained on the Germplasm Resources Information Network (GRIN). A diskette version (PCGRIN) of the database is available at no charge. PCGRIN contains passport and evaluation data and is updated annually. To receive PCGRIN on disk contact :

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Table 1. Growth of the *Helianthus* collection (NCRPIS, 1989-1993).

YEAR	CULTIVATED ACCESSIONS	WILD ACCESSIONS	TOTAL NUMBER OF ACCESSIONS
1989	1262	1867	3129
1990	1360	1869	3229
1991	1371	2009	3380
1992	1435	2149	3584
1993	1445	2163	3608

Table 2. The number and percentage of *Helianthus* packets distributed to the domestic public, the domestic private, and the foreign user communities (NCRPIS, 1989-1993).

	DOMESTIC PUBLIC		DOMESTIC PRIVATE		FOREIGN		
YEAR	NUMBER	%	NUMBER	%	NUMBER	%	TOTAL
1989	2111	39	491	9	2776	52	5378
1990	657	25	1250	47	745	28	2652
1991	1186	46	620	24	770	30	2576
1992	1764	68	411	16	402	16	2577
1993	700	30	307	13	1352	57	2359
Cumulative Total 1989-1993	6418	41	3079	20	6045	39	15542

ENTOMOPATHOGENIC NEMATODES - AN OVERVIEW

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Introduction:

Entomopathogenic nematodes are obligate parasites primarily infecting members of the Class Insecta, although other invertebrates are being added to the host range as evaluations broaden in scope. Several families of Nematoda are known to rely on insects as hosts for reproduction and, therefore, survival. Two families, the Steinernematidae and Heterorhabditidae, are the main focus of current research efforts geared towards biological control applications. Several other taxa have been studied as host:parasite models; however, their efficacy is questionable on an agricultural scale, primarily due to difficulties in production (*i.e.*, rearing) and narrow host range.

Several strains of species in the Steinernematidae and Heterorhabditidae have been selected and even improved through planned matings, such that knowledge of these groups surpasses that of other nematode entomopathogens. The genera *Steinernema* and *Heterorhabditis*, of the families Steinernematidae and Heterorhabditidae, respectively, are commercially available and will be the focus of this presentation.

Symbionts and Reproductive Life Cycle:

Both genera, *Steinernema* and *Heterorhabditis*, share many common features in their biology with some key differences. Upon entry into a host insect through mouth, anus, spiracles or (less commonly) wounds, the infective juvenile (IJ) stage of these nematodes proceeds to the hemocoel to initiate host death via immunosuppression and release of symbiotic bacteria from the gut and/or pharynx of the IJ.

The symbiotic bacteria housed in the intestine of the IJ are required for successful infection (*i.e.*, ensuing septicemia), antibiosis with respect to native microfloral suppression, and provision of a nutritive environment for the development of adult nematodes. Gnotobiotic nematodes may kill their host; however, reproduction requires the influence of the gram-negative symbionts in the degradation of host tissues. Hydrolyzed host components supply the necessary nutrition for development of adult nematodes, progeny, and ultimate release of the nonfeeding IJ; these are released upon exhaustion of nutrients to go in search of another host. *Xenorhabdus* species are found within *Steinernema* in a species-specific association. A single species, *Photorhabdus luminescens*, is associated with members of *Heterorhabditis*, although strain specificities exist to varying degrees.

The two families differ primarily in reproductive behavior by the presence of hermaphroditic adults in *Heterorhabditis* and their absence in *Steinernema*. Both shift between oviparous and ovoviviparous production of progeny, resulting in the development of three juvenile stages prior to final egress of IJ.

Applications to Biological Control:

Mass-fermentation technology has allowed the production of several species and their respective strains in quantities sufficient to make field applications economically feasible. Many steinernematid species can be produced for approximately $0.10 to $0.15 per million IJ. Rates vary with target insect hosts and crop species, but many field-row crops are treated with 1 X 10^9 to 3 X 10^9 IJ/A. Actual area covered is determined largely by precision of application and position of target pest (*e.g.*, only within rows; at base of orchard species; within high-value container stock, etc.). Hence, the cost/A varies considerably with need.

Primarily, nematodes have been applied to soil-borne insects due to problems of UV sensitivity and desiccation in above-ground (*i.e.*, leaf-feeding pests) applications. Many strains of entomopathogenic nematodes have broad host ranges and are, therefore, a potential biopesticide for many insect pests. No definitive demonstration of harmful effects on beneficial insect populations have been cited; however, the quantity of research needed in this area is lacking.

Inundative release of IJ is one approach to control a pest within a localized area over the short term. Benefits may include rapid decimation of the feeding populations; however, 'knock-down' may not be as dramatic as with chemical insecticides. Another approach to control is an inoculative release of entomopathogenic nematodes, which relies on survival of a remnant host population to ensure reproduction of sufficient nematodes to combat deviations in host numbers. The latter method is gaining wider acceptance as integrated pest management (IPM) strategies are assessed.

Various formulations of nematodes are available for packaging, storage and application. The most common method of shipping and storage is to embed the IJ in an alginate matrix. Dissolution of nematodes is accomplished by imbition of water or a citrate-EDTA mixture. Other less common packing and application materials include peat, vermiculite, clay and charcoal. Newer methodologies include granular, flowable formulations, which will enter the market this year.

Considerations in selection of nematode strains for targeting specific insect pests require an *a priori* knowledge of at least the general biology (*i.e.*, life cycle, behavior, feeding site) of the potential host. With a soil-borne insect, depth within the soil profile, emergence-development dates, soil type and mobility of the host are all significant factors.

Heterorhabditis bacteriophora strains tend to be mobile with respect to dissemination from the point of application, whereas *Steinernema carpocapsae* often remains predominantly near placement sites. Different strategies of nematode species, such as the 'wait and ambush' nature of many steinernematids or the 'searcher-cruiser' movements of *S. glaseri* or *H. bacteriophora*, will vary in their infection rate in large part due to behavioral defense responses of insect hosts. IJ often sense CO_2 release or various kairomone clues to locate hosts; evolutionary selection has apparently provided for alteration in CO_2 release patterns or secretion of 'repellants' (*e.g.*, frass of sciarids) as avoidance responses.

Insects may also preclude entry of nematodes by preening, sensitive oral receptors and regurgitation, and sieve plates covering spiracles (as in some sciarids). Additionally, certain stages in the host life cycle may be more susceptible to nematode infection and, therefore, targeting such a stage (*e.g.*, larva, adult) may result in greater control than application to the destructive or most evident stage.

Soil moisture and structure are critical to the success of nematode applications. IJ may enter a resting state, similar to anhydrobiosis as noted in aphelencoid nematodes, when they undergo slow desiccation. Rapid decreases in relative humidity (RH) will often cause death of IJ, however, even shallow watering following application to soil can aid greatly in survival. Conversely, soils near field capacity may inhibit nematode movement and eventually result in the demise of IJ if the duration of standing or free-water is greater than a couple of days; clay soils are especially prone to this phenomenon. Water bound to soil particles by capillary, adsorptive and absorptive forces may provide sufficient RH to ensure nematode survival due to reduced flux as compared to soil surface or atmospheric changes with time.

Soil particle size, type and percent organic matter also influence IJ movement and survival. Nematodes utilize sinusoidal movements, particle to particle 'leaping' and other soil macrobiota as means of active dispersal. Flow or irrigation water, rain, wind and host movement prior to death are passive forces which can influence spread of IJ within the soil profile. Hence, the structure of the soil (*i.e.*, pore sizes of interstitial spaces) potentiates the degree to which these active and passive mechanisms mediate dispersal and host contact. Some pesticides, such as the insecticide-nematicide oxamyl, can even enhance the movement of IJ via behavioral stimulation at typical field concentrations.

A variety of chemical pesticides have been shown to influence nematode survival via their inherent toxicity. These include herbicides, nematicides and insecticides of several chemical structural groups. Chloropyrifos (Lorsban), applied at recommended field application rates, has acted both synergistically and without apparent influence on nematodes in several cropping situations. Others, such as aldicarb (Temik) or carbofuran (Furadan), have shown significant toxicity to IJ in some applications. The compatibility of pesticides used in combination with entomopathogenic nematodes depends on rate of application, residue stability, time of application relative to release of IJ, the species of nematode, soil interactions, site of pesticide application, and presumably other factors which are presently unknown. Unfortunately, no predictive means exist to assess potential influences or interactions between chemicals and nematodes.

Several miscellaneous factors also contribute to the ultimate influence of IJ on host populations. As host behavior varies with species, so does the behavior of nematode species and even strains. Most heterorhabditids are active dispersers, UV susceptible, and do not store as well as steinernematids. Additionally, most heterorhabditids appear to predominate in downward movement in soil and are of the 'searcher-cruiser' type. The smaller steinernematids, such as *S. carpocapsae*, tend to remain close to the site of application and are of the 'ambusher' type. *S. glaseri*, a larger species, is more mobile than others in the family, however; it tends to move up in soil columns when placed within the profile, is a 'searcher-cruiser' type, and is capable of significant active dispersal. Unfortunately, this species is also poor with respect to retention of bacterial symbionts and costs approximately 10 times more than most other steinernematids.

Sugarbeet Root Maggot Biological Control:

One of the primary efforts in the Sugarbeet Research Unit at Fargo is the application of entomopathogenic nematodes for control of the sugarbeet root maggot (SBRM), *Tetanops myopaeformis*, a dipteran of economic importance.

Laboratory assays with six strains of *Steinernema spp.* and four strains of *H. bacteriophora* demonstrated that third instar SBRM larvae were susceptible to IJ co-incubated at rates similar to field applications. Strains and species varied in their respective abilities to infect and reproduce within SBRM larvae; however, percent mortality overall was low (4-9%) following 72-h incubations in sand. The reasons for this are unclear, as incidents of infection typically yielded reproduction and egress of thousands of IJ per larva. We noted in some experiments that initial infections, evidenced as enlarged, adult nematodes within the host, resulted in no further reproduction of nematodes and dissection of cadavers revealed only a deliquescing liquid mass devoid of nematodes. The presence of host microflora in this insect is well documented, and we speculate that antibiotic production by endogenous bacteria may have nullified the septicemic conditions required for nematode nutrition.

When adult SBRM (flies) were co-incubated with the same six steinernematid strains, however, the susceptibility of the adult stage was found to be much greater. Flies housed in laboratory containers with IJ for as little as 2 h were infected, deceased within 6 to 24 h post-incubation (42%), and demonstrated reproduction of nematodes with egress of IJ at 4 to 7 days post-incubation. Longer periods of co-incubation resulted in an increased mortality of flies (*e.g.*, 6 h - 70%, 21 h - 77%, 36 h - 100%).

Given the differential between larval and adult susceptibilities, we have planned a lure/attractant study to determine if significant numbers of adults can be infected following visitation to milk-carton traps housing IJ in a sand:polyacrylamide mixture. Preliminary trials have demonstrated the infection of adults under field conditions, although due to windy, rainy weather, sufficient numbers were not attainable for statistical analysis. Evaluation of 'trap' design and chemical lures is planned for this season.

Red Sunflower Seed Weevil:

Preliminary experiments with the red sunflower seed weevil, *Smicronyx fulvus*, indicated the potential for use of entomopathogenic nematodes in a control program. Laboratory co-incubations of *S. feltiae* and *S. carpocapsae* resulted in infection of *Smicronyx* larvae with subsequent egress of IJ. Due to the small size of the experiments, it is not possible to provide quantitative assessment of control potential of this pest using steinernematids. However, as with all field applications, the only true test of efficacy is a controlled study of strain abilities within the typical cropping system.

Again, soil type, timing and rate of application, strain selection, chemical compatibilities and other factors will all influence the success of host population control. Since this insect spends significant time in the soil, it is more likely to be accessible to control as a larva, as opposed to targeting adults in the hostile environment of the seed head. Desiccation and UV

susceptibilities have limited 'aerial' (*i.e.*, above ground) applications, although newer formulations of IJ with osmo- and UV-protectants may moderate this in the near future.

Miscellaneous Considerations in Application of Entomopathogenic Nematodes for Biological Control:

In addition to factors cited above, several others may have varying degrees of influence on the choice of selecting entomopathogenic nematodes or other measures for pest control. Some of these are biological in nature, but in many instances non-biological considerations, such as economics and grower acceptance, can be equally important.

Several commercial concerns in the U.S. and abroad currently produce nematodes for insect control. The size of these firms varies from small, cottage industries to major corporations. Although entomopathogenic nematodes have been tested with some regularity for over 60 years in the U.S., they have only come to the forefront of biological control in the past decade or less. Hence, the acceptance by growers, IPM specialists, consultants and the public will likely require significant education to ensure a market exists for survival of manufacturers distributing them.

Comparisons with chemical control measures have been mixed, with equivalent control documented as well as insufficient control. Modification of application methods, novel strains of nematodes, use of chemical modifiers (*e.g.*, stimulants, anti-desiccants, etc.), and improved storability will all likely play a role in the overall success of entomopathogenic nematodes in agriculture. Those expecting rapid death of target insects, as found in many chemically-based controls, may ultimately find fewer chemicals available for registered use and, hence, be positioned to examine biologically-based methods giving moderation of host population numbers over a longer term. Inoculative-release practices have gained interest and support from those successful programs which rely on augmentation from beneficial insects and natural epizootics in addition to direct application of biopesticides.

Cost of IJ and any equipment purchases needed for application are, of course, the bottom line for many growers. If their cost is not in line with crop-value losses, their use is prohibitive and alternatives will be sought. Similarly, the cost of IJ production can vary with species (*e.g.*, *S. glaseri* is expensive, *S. feltiae* is inexpensive) and thereby influence the availability of strains, regardless of any biological benefits. The number of producers of entomopathogenic nematodes and, therefore, the total number and types of available strains, will also be determined by economic considerations. Should the widespread use of nematodes in bio-control be stayed, the number of producers will undoubtedly be fewer.

With the number of documented successful applications of entomopathogenic nematodes, alone or in combination with chemical pesticides, ever increasing, the utilization of these parasites for control of pests will likely increase also. The safety and low environmental impact of this biopesticide are attractive features that will be of increased significance following a general reduction in registration of broad-spectrum insecticides.

Suggested Readings:

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NATURAL CONTROL OF SUNFLOWER INSECT PESTS BY INDIGENOUS PARASITOIDS

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The natural control of insect pests is influenced by a number of factors, including: weather; food availability; spatial or territorial requirements; interspecific and intraspecific competition; and the actions of natural enemies. Due to these factors and especially the natural enemies, which operate in a density dependent manner, less than 1 to 2 percent of phytophagous insects ever become serious pests. Natural enemies include predators, parasitoids, and diseases. Studies in different cropping systems (alfalfa, grain, collards, brassicas) have shown that from 47-67% of the species of arthropods present are natural enemies. Natural enemies may be utilized in the following ways to control pest species: 1. importation and establishment of exotic species; 2. augmentation of established species; or 3. conservation of species through manipulation of the environment.

The conservation of natural enemies in a crop agroecosystem is important in maintaining pests below levels that cause economic damage. The naturally occurring or indigenous natural enemies prevent many plant-feeding insects from achieving pest status. The conservation of these natural enemies allows them to operate at their full potential. Manipulating the environment to eliminate or mitigate adverse factors, such as pesticides, can effectively conserve the natural enemies present. The use of pesticides results in the destruction of the target pest as well as the natural enemies that are present. The effective conservation of natural enemies depends on the following approaches: understanding of the agroecosystem; the use of selective pesticides; the use of the least disruptive formulation of the chemical; application of the insecticide only when necessary and based on sound economic injury levels of the pest; and pesticide application at the least injurious time or place.

Sunflower is native to North America. Insects associated with sunflower (*Helianthus* spp.) have evolved with the plant for centuries and many have moved to the cultivated crop to feed and develop. These include phytophagous species, pollinators, and natural enemies. Although hundreds of insects have been recorded from sunflower, only a small number have achieved pest status. Indigenous natural enemies have been a significant factor in preventing many insects from becoming economic pests. The insect species that have become pests are also subject to attack by numerous natural enemies, including predators, diseases and parasitoids. The following discussion reports studies of parasitic species associated with the sunflower

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stem weevil, *Cylindrocopturus adspersus* (LeConte) (Coleoptera: Curculionidae), and the red and gray sunflower seed weevils (Coleoptera: Curculionidae), *Smicronyx fulvus* LeConte, and *S. sordidus* LeConte, respectively.

Sunflower Stem Weevil - The sunflower stem weevil is attacked by both egg and larval parasitoids. Hymenoptera recovered from overwintering larvae include: *Tetrastichus ainsliei* Gahan (Eulophidae); *Mesopolobus* sp. (Pteromalidae); and *Nealiolus curculionis* (Fitch) (Braconidae). The latter species is the most prominent of the larval parasitoids attacking the weevil. The eggs of the weevil are attacked by *Anaphes pallipes* (Ashmead) (Hymenoptera: Mymaridae).

Nealiolus curculionis is a univoltine, solitary, endophagous larval parasitoid of the sunflower stem weevil in both cultivated and native sunflower. This parasitoid represented 96% of all parasitoids attacking the weevil from 1980 to 1991. Adult parasitoids are active in the field from late June to late August. Eggs are deposited in early instar weevils feeding within the sunflower stalk. The immature parasitoids overwinter within diapausing weevil larvae in the sunflower stalk.

Parasitization of overwintering sunflower stem weevil larvae varied from 5 to 32% from 1980 to 1991 (Fig.1). Results indicated that overall parasitization has increased from levels reported in the late 1970's and early 1980's. Except for 1989, and the period 1986 to 1988 when no data are available, parasitism of stem weevil larvae by *N. curculionis* has averaged 27% since 1983, while stem weevil densities have varied from 6 to 29 larvae per stalk. The rate of parasitism showed an increase from 1981 to 1983 as the population of weevil larvae decreased from 108 to 24 larvae per stalk. But, the consistent rates of parasitism (Fig.1) compared with the variable field densities of adult parasitoids suggest that *N. curculionis* effectively forages for hosts under varying host population densities. The ability of the female parasitoid to locate and attack hosts is of paramount importance to the success of a given parasitoid population. The parasitoid appears to be a consistent mortality factor in the population dynamics of the sunflower stem weevil in cultivated sunflower.

Studies at both Prosper and Carrrington, North Dakota, in 1991, revealed decreases in stem weevil larval densities in stalks as the planting date was delayed, but no significant difference in rates of parasitization (Figs. 2 & 3). Although *N. curculionis* was not recovered from larvae in stalks of the third planting date, this was probably due to the extremely low number of weevil larvae present. Parasitism of overwintering stem weevil larvae showed a numerical difference among dates, but the differences were not statistically significant. This was probably due to the variability in parasitism of larvae in different stalks within each planting date. The results revealed that the parasitoid was active and capable of attacking larvae of the sunflower stem weevil in sunflower from different planting dates.

Comparison of densities of weevil larvae showed significant differences among different sunflower lines (Fig. 4). Numbers of larvae varied from 17 to 59 per stalk showing that some lines have potential resistance to infestation by the stem weevil. Parasitization rates also varied among lines (6 to 34%), with high parasitism in some with low weevil numbers. Parasitism also varied among some lines with similar weevil densities. The mechanisms that

influence parasitism of weevil larvae among different sunflower lines need to be investigated.

Nealiolus curculionis also attacks sunflower stem weevil larvae in native *Helianthus annuus* L. In eastern North Dakota and western Minnesota about 19% of the larvae in *H. annuus* from three locations were parasitized (Fig. 5). The parasitoid showed evidence for good searching ability, since populations of stem weevil larvae were quite low within these stalks. Parasitoids were not recovered from weevil larvae in stalks of *H. petiolaris* Nuttall, *H. maximiliani* Schrader, or *H. tuberosus* L., probably because of the small populations of hosts present.

Sunflower Seed Weevils - Larvae of both the red and gray sunflower seed weevils are attacked by hymenopteran parasitoids. The dominant species appear to be specific to each weevil species parasitized. The braconid wasps, *Urosigalphus femoratus* Crawford and *Triaspis aequoris* Martin have been reared from overwintering gray and red sunflower seed weevil larvae collected from cultivated sunflower at Leonard and Prosper, North Dakota, respectively. These were the only parasitoid species recovered from overwintering red and gray sunflower seed weevil larvae in cultivated sunflower collected at nine sites throughout North Dakota in 1991 and 1992. They are both solitary endoparasitoids that overwinter within the seed weevil larva in the soil, destroying the larva before it can pupate the following summer.

Dissection of red sunflower seed weevil larvae collected from cultivated sunflower at Prosper, North Dakota, from 1988 to 1993 has shown an increase in parasitism (4 to 50%) by *T. aequoris* over time (Fig. 6). The parasitoid female deposits eggs singly in the eggs and early instar larvae of the weevil. The parasitoid larva develops within the host, but does not kill the weevil larva until the following season.

The emergence patterns of both the adult parasitoid, *T. aequoris*, and the weevil host were monitored at Prosper and Carrington, North Dakota. Emergence for both the parasitoid and host were earlier at Carrington than Fargo, but at both locations, most of the parasitoids emerged over a one month period, beginning in early August (Fig. 7). *Triaspis aequoris* had generally a more rapid emergence than its host, the red sunflower seed weevil.

Parasitization rates of red sunflower seed weevil larvae recovered from three different dates of planting were compared at Prosper, North Dakota, in 1992. Larvae were dissected to determine the number of larvae attacked. The results showed that *T. aequoris* was active throughout the season, parasitizing red sunflower seed weevil larvae in heads from all three dates of planting (Fig. 8). Parasitization rates from both the first and third planting dates were almost identical. The reason for the decline in parasitism of larvae from the second date is unclear.

The parasitoid fauna attacking the red and gray sunflower seed weevils in native *Helianthus* was more diverse than in cultivated sunflower. Seed weevils in the larval stage could not be identified to species, therefore the results on species of parasitoids were from one or both seed weevils. A total of five species of parasitoids were reared from overwintering weevil larvae collected from different sunflower species in South Dakota, Kansas, Nebraska,

Wyoming, Colorado, Montana, and North Dakota in the fall of 1991. The following braconid wasps were recovered: *T. aequoris* in larvae from *H. annuus* and *H. petiolaris*; *N. curculionis* in larvae from *H. annuus*, *H. nuttallii*, and *H. maximiliani*; *N. rufus* (Riley) in larvae from *H. annuus* and *H. petiolaris*; and *U. femoratus* in larvae from *H. annuus*, *H. petiolaris*, *H. nuttallii*, and *H. maximiliani*. A pteromalid wasp, *Eutrichosoma mirabile* Ashmead, was reared from larvae collected in heads of *H. annuus* and *H. maximiliani*. Further research is needed to increase the diversity of parasitoids attacking weevil larvae in cultivated sunflower.

Natural enemies are an important component in the crop agroecosystem. They prevent many phytophagous insects from becoming pests and causing economic damage to the crop. Predators, parasitoids and diseases also attack insect pests and the degree of control they exert can be managed by using tactics that conserve these natural control agents. Sunflower pests are also susceptible to attack by a variety of natural enemies, including different species of parasitic wasps. An understanding of the species present in both native and cultivated sunflower, rates of parasitism, biology, population dynamics, and influence of different control methods is important in developing ways to increase the impact of these parasitoids as biological control agents of sunflower insect pests.

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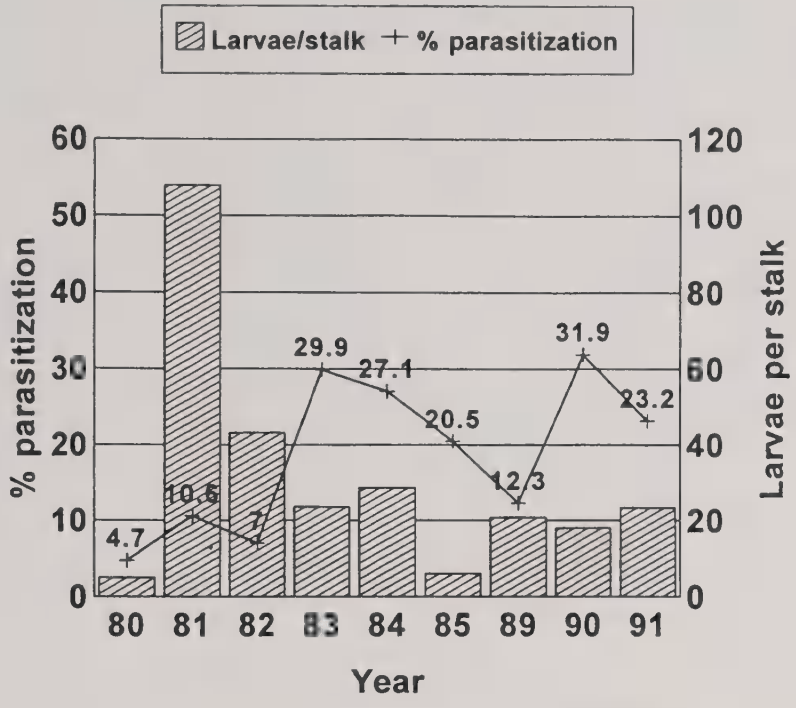

Fig. 1. Number of sunflower stem weevil larvae in sunflower stalks and percentage parasitization by *Nealiolus curculionis* from 1980 to 1991 in North Dakota.

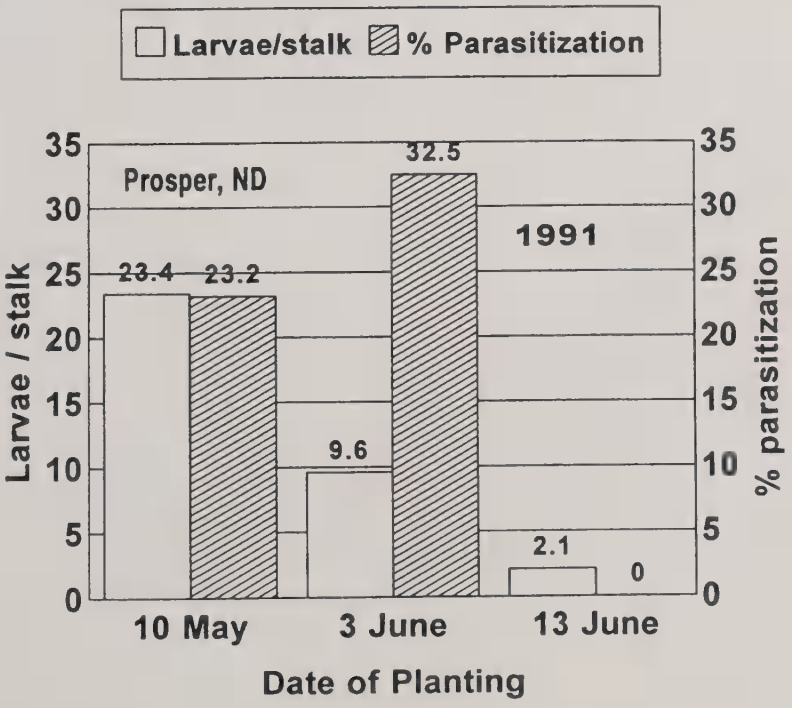

Fig. 2. Number of sunflower stem weevil larvae in sunflower stalks and percentage parasitization by *Nealiolus curculionis* from three different planting dates, Prosper, North Dakota, 1991.

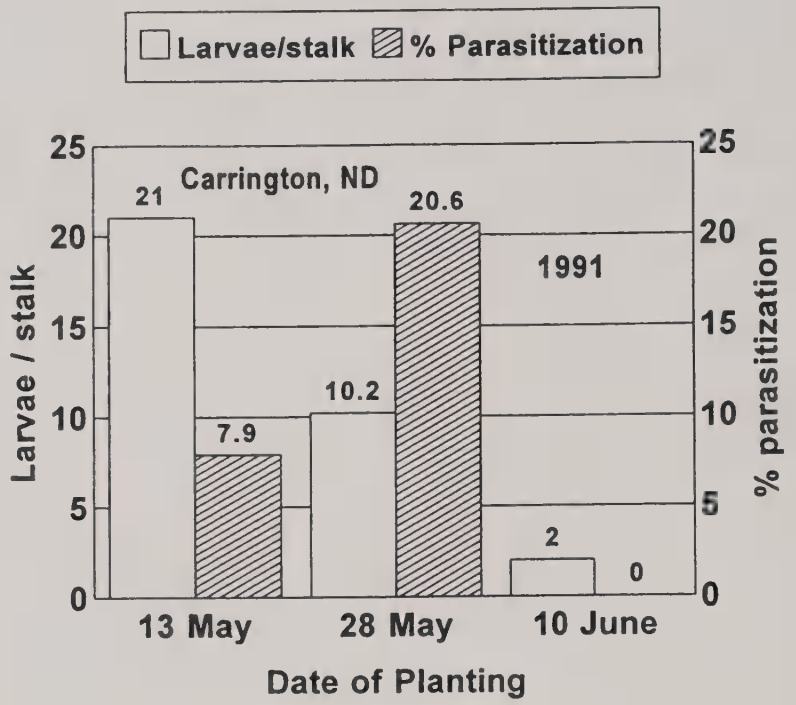

Fig. 3. Number of sunflower stem weevil larvae in sunflower stalks and percentage parasitization by *Nealiolus curculionis* from three different planting dates, Carrington, North Dakota, 1991.

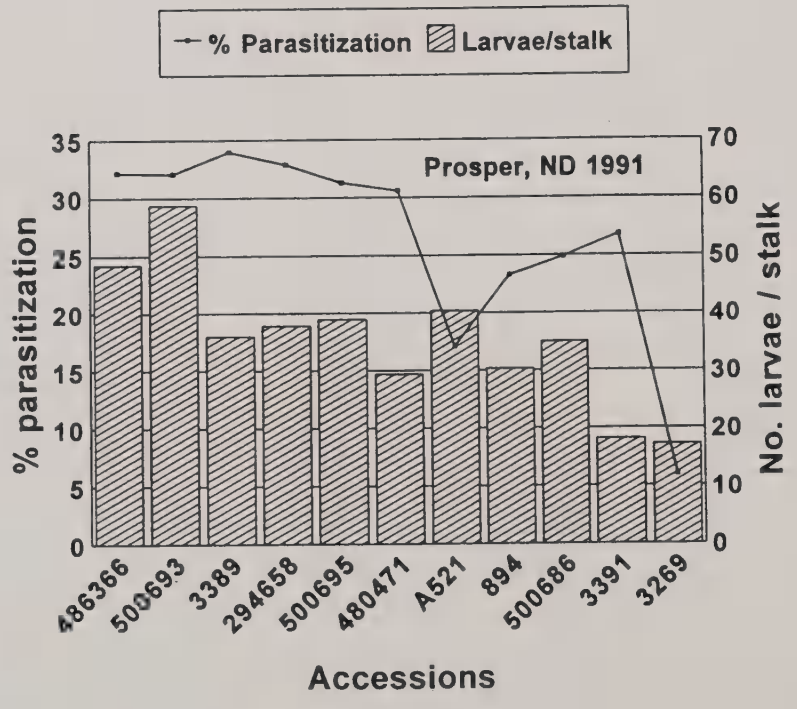

Fig. 4. Number of sunflower stem weevil larvae in sunflower stalks and percentage parasitization by *Nealiolus curculionis* in selected sunflower lines at Prosper, North Dakota, 1991.

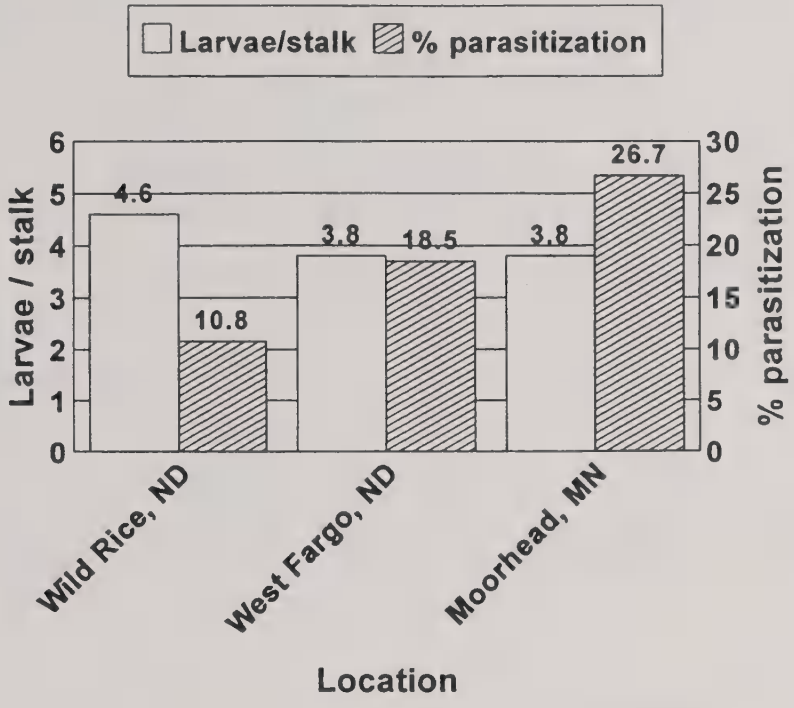

Fig. 5. Number of sunflower stem weevil larvae and percentage parasitization by *Nealiolus curculionis* in native *Helianthus annuus* at two sites in eastern North Dakota and one in western Minnesota, 1988.

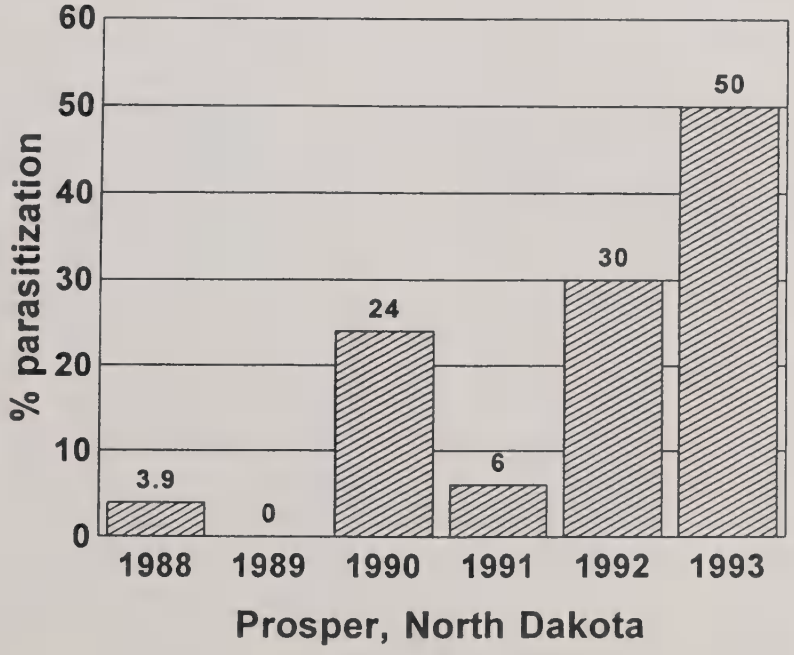

Fig. 6. Parasitization of red sunflower seed weevil larvae by *Triaspis aequoris* from 1988 to 1993, Prosper, North Dakota, based on dissected larvae.

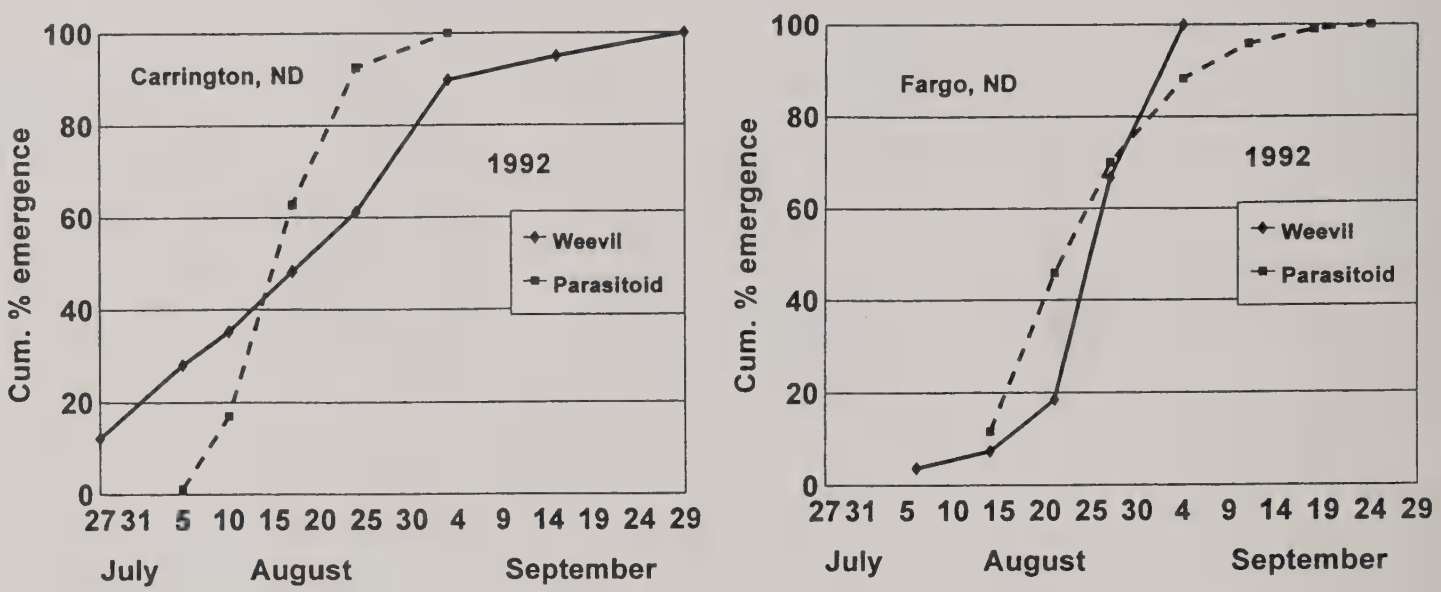

Fig. 7. Emergence pattern of adult red sunflower seed weevil and its parasitoid, *Triaspis aequoris*, at Prosper and Carrington, North Dakota, 1992.

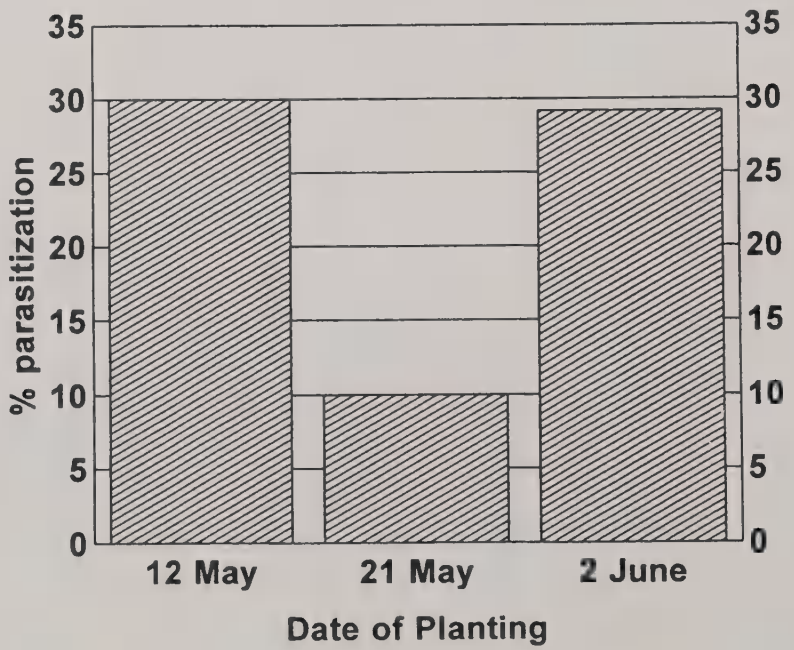

Fig. 8. Parasitization of red sunflower seed weevil larvae by *Triaspis aequoris* from three different planting dates, Prosper, North Dakota, based on dissected larvae.

FEEDING BEHAVIOR OF ADULT SUNFLOWER SEED WEEVILS ON CULTIVATED SUNFLOWERS

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Infestation by the sunflower seed weevil complex has been a major problem in sunflower production areas in the Northern Great Plains. Occurrence of the gray sunflower seed weevil, *Smicronyx sordidus* LeConte on commercial sunflowers relative to the occurrence of the red sunflower seed weevil, *S. fulvus* LeConte, was estimated to be 1 to 20 or 30, respectively (Satterthwait, 1946). The former is more common in the southern latitudes of the United States than the latter (Rogers, 1988).

Adults of the gray sunflower seed weevils begin emerging on 1 July in North and South Dakota, with 50% emergence by 10 August (Gednalske and Walgenbach, 1984a). They feed on stems and petioles before the bud stage. Eggs are laid on the distal end of unopened florets before the bloom stage. Larvae migrate down the corolla, penetrating the developing achene at the point of attachment between the corolla and the ovary, and feed on the embryo at the achene base. The infested achenes lack an embryo at maturity in contrast to uninfested achenes (Brewer, 1991).

On the other hand, adults of the red sunflower seed weevils first appear on commercial sunflowers during the last week of June in North Dakota (Schneiter and Miller, 1981). Adult population reaches its peak at the R5.5 to R5.9 substages of plant development (Oseto and Korman, 1986). Adults feed on stem and leaf tissue before the reproductive stage of the plant and finally move to the inflorescence to feed on pollen or florets at anthesis. Eggs are laid between the pericarp and the embryo of the developing achenes during anthesis. However, pollen depletion and hardening of achene walls prevent further oviposition and result in movement of the weevils into other pollen-producing plants in the field. About 2/3 of the embryo is left after feeding is completed.

At the prebloom stage of sunflowers, population density of the gray sunflower seed weevils increases, but declines after bloom or anthesis occurs (Byers, 1987). In contrast, the red sunflower seed weevils become more abundant at bloom or anthesis stage.

The goal of this study was to: a) determine the type of tissue consumed by adults of *Smicronyx fulvus* ; and b) describe the effect of feeding at different plant growth stages on the maturation of the female reproductive systems of *S. fulvus* and *S. sordidus* .

Materials and Methods

Determination of the type of tissue consumed by adult *S. fulvus* LeConte.

One experiment was conducted in the laboratory by confining adult sunflower seed weevils in petri dishes measuring 15.5cm dia x 2.5cm deep. Weevils in each petri dish were provided with freshly excised florets (T1), leaf (T2), or bracts (T3). The set-up was held in a growth chamber at 28°C, 55%RH, and 15:9hrs (L:D) cycle for 6 days. The number of feeding punctures by males, females, and paired (male+female) weevils on each substrate was compared. A follow-up experiment was also conducted to determine the preference of each sex (male or female) to different sunflower plant parts. This was conducted using newly-emerged adults confined individually in a clear rectangular container (36cm W x 14cm H) with florets, leaf and bracts placed separately within the container. The time (resting+feeding) spent by each individual on each plant part was recorded continuously for a period of 24hrs in the laboratory.

Effect of feeding at different plant growth stages on the maturation of the female reproductive systems of *S. fulvus* and *S. sordidus* LeConte

The experiment was conducted in the greenhouse by confining 20 pairs (male and female) of newly-emerged adults on the capitulum, including the uppermost (3-4) leaves, using delnet bags beginning at the R1 stage of plant growth. Treatments were represented by 2 pairs of weevils collected from each capitulum at 2 day intervals until the anthesis stage (R5.1-5.3). The effect of feeding at various plant growth stages on egg development or maturation was described and compared.

The same set-up as above was conducted simultaneously except the weevils were introduced at the anthesis stage (R5.1-5.9) to determine the influence of pollen availability on female reproductive maturity. In the laboratory, an equal number of weevils were confined in petri dishes and provided with moist filter paper to serve as controls. Two pairs of weevils were taken from each capitulum and petri dish at 2 day intervals until the plants in the greenhouse reached the R6-7 growth stage. Egg maturation of females from the capitula was compared with those from the petri dishes.

Results and Discussions

Type of tissue consumed by adult *Smicronyx fulvus* LeConte

It was observed that adults utilized almost all parts of the sunflower plant for food. They fed on tissues of the young, succulent stems, leaves, bracts, petals, and florets. However, among the selected plant parts used, florets appeared to be the most preferred plant part for feeding by females (Table 1). And males fed equally on leaves and florets. The majority of the male

weevils fed on the base of the corolla where the nectary is located, while majority of the females fed on both the base of the corolla and developing achenes. This shows that since the immature stage has evolved or adapted to feed on the capitulum, particularly the developing seeds, adults are most likely to be adapted to obtain adequate food in plant structures adjacent to the larval niche.

On the other hand, feeding punctures by paired weevils on florets suggested that the presence of males did not alter the feeding response of females. The suitability of florets for food by both male and female weevils is associated with the longer time spent by each individual on florets. Under choice conditions, the weevil's time spent on the florets was characterized by resting and feeding states while no feeding was observed during their time on the bracts or leaves. However, the feeding observed on leaves and bracts indicate that weevils would be forced to feed on these plant parts for survival until the most preferred food substrate is available.

Effect of feeding at different sunflower growth stages on the maturation of the female reproductive systems of *S. fulvus* and *S. sordidus* LeConte

A. *Smicronyx fulvus* LeConte

Examination of the female ovaries revealed that at the prebloom stages, R1 through R4, oocyte development in the ovarioles was not followed by ovulation, the passage of mature oocytes into the oviduct. Instead, oocyte resorption or oosorption was evident especially at 4 and 8 days after exposure when plants were at R2-4 growth stage. Resorption was characterized by shrinkage of oocytes which ultimately result in a collapse of the whole follicular epithelium. The process of resorption provides a mechanism for making optimal use of available nutrient so that only eggs with adequate amount of yolk are laid to insure survival (Chapman, 1982). At the anthesis or bloom stage, 16 days after exposure, eggs were found in the lateral oviduct of the female. These observations clearly showed that lack of or poor-quality food for the adult female at the prebloom stages of sunflower development may cause a delay in the onset of egg production, an increase in oosorption, a possible cessation of oogenesis, and to some extent a reduction in the rate of egg production.

Results also revealed that when weevils were exposed to blooming sunflowers, mature oocytes were developed in the ovariole as early as 2 days after exposure. Two days later, eggs were already found in the lateral oviduct ready for deposition. On the other hand, when weevils were provided only with water, oocyte resorption was observed after 2 days. And no eggs could be found in the lateral oviduct even after 12 days. These results further indicate the capability of newly-emerged adults to produce eggs when good-quality food and the proper substrate for oviposition is readily available. Leaves and other plant structures appear to be unnecessary, since egg development occurred when weevils were restricted to feed on the capitulum. Thus, sunflower pollen or carbohydrates and/or

amino acids in the nectar are probably essential to produce and provision eggs.

B. *Smicronyx sordidus* LeConte

Unlike the female red sunflower seed weevils, egg development in female ovaries of the gray sunflower seed weevils was evident as early as 4 to 6 days after exposure when plants were at R2 growth stage. The eggs were found in the lateral oviduct of the female ready for deposition. However, when newly-emerged adults were introduced immediately to blooming sunflowers, no eggs were produced even after 12 days of exposure. These results suggested that the gray sunflower seed weevils do not require pollen for egg development. Tissues of young, succulent leaves, stem, and bracts seemed adequate for maturation of the female reproductive system of the gray sunflower seed weevils as opposed to the red seed weevils. The capability of this species to produce eggs before anthesis stage is probably the reason why the population normally declined during anthesis stage in the field.

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Table 1. Average time spent and feeding punctures by adult red sunflower seed weevils on different sunflower plant parts.[a]

Treatment	Time (min)[b]		Feeding Puncture		
	Male	Female	Male	Female	Pair
Florets[c]	824 a	823 a	10 a	13 a	32 a
Leaf	3 b	287 a	3 ab	1 b	9 ab
Bracts	171 a	3 b	1 b	0 b	0 ab

[a] Means in a column followed by the same letter are not significantly different (P>0.05) (HSD); data based on 5 replications each

[b] Based on 24hr observation

[c] Ray flowers were included only in the 24hr observation but not in the feeding puncture experiment

Economic Injury Levels for the Red Sunflower Seed Weevil (Coleoptera: Curculionidae) Infesting Oilseed Sunflower

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THE RED SUNFLOWER seed weevil, *Smicronyx fulvus* LeConte, is the most serious pest of sunflower in the Northern Great Plains (Oseto & Korman 1986, Charlet et al. 1987). Females oviposit in the achenes from mid-July to mid-August. Larvae feed on the kernel. Most larvae drop to the soil before harvest, and overwinter in the soil. Some larvae remain in the achenes after harvest, especially during a cool growing season (Oseto & Braness 1979). Management options for the weevil are limited. Early planting minimizes weevil damage but also may reduce seed weight and oil content (Oseto et al. 1987). Other management strategies, such as plant resistance and trap cropping, are currently under study. Currently, sunflower growers rely on insecticides for economic control of the weevil (Oseto & Burr 1990).

Establishment of an efficient insecticide control program requires knowledge of an economic injury level (EIL) (Smith 1968). The EIL is defined as the lowest population density that will cause economic damage (Stern et al. 1959). Control measures should not be carried out unless the pest population is very likely to exceed the EIL (Poston et al. 1983). Although an EIL for the weevil was determined by Oseto and Braness (1980), several questions remained unsettled. First, although oil content loss resulted from larval infestation (Braness 1978, Oseto & Braness 1980), it was not considered in the calculation of the EIL. Second, a mean of 19.9 larvae produced by a female (Oseto & Braness 1979) was used to construct the EIL. In single plant cage tests where newly emerged weevils were placed on sunflower heads at plant stages of R5.6-5.8 (Schneiter & Miller 1981), the females produced an average of 18.1 larvae (Oseto & Braness 1979). Because females require pollen for 5 d to mature eggs (Korman & Oseto 1989) and plant stages after R6.0 are not suitable for oviposition by the weevils (Oseto & Braness 1979), the length of time available for weevil oviposition was too short in the single plant cage test; therefore, the female reproductive rate was reduced. We believe that females would produce more larvae under natural conditions than the number of larvae indicated by Oseto and Braness (1980). Third, for the EIL value to have utility in a pest management program, the relationship between the number of adults counted at different plant stages and the subsequent number of damaged achenes must be understood. There has been no study to specify the plant stages on which adult counts should be taken to calculate the EIL, although adult counts before plants reach stage R5.4 (McBride et al. 1992) and at the R5.5 stage (Oseto & Korman 1986) are recommended .

The objectives of this study were to determine the weight loss due to larval feeding and the relationship between the number of adults counted at different plant stages and the subsequent number of damaged achenes, which then were used to redefine the EIL.

Materials and Methods

Three North Dakota fields were surveyed for adult weevil populations and achene damage. Fields were at the North Central Research and Extension Center at Minot (1992 and 1993) and Research Farm at Prosper (1993). Field size ranged from 0.4 to 4 ha. Sunflower hybrid 'DO 855' was planted at Minot in 1992. Sunflower hybrid 'Interstate 894' was planted at Prosper and 'Northrup King 265' at Minot in 1993. Plant population was about 46,000 plants per ha. Preplanting applications of herbicides were applied and hand removal of weeds was used as needed. Insecticides were not applied to any of the fields.

Sampling for adult weevil population was initiated at bud stages (mostly R3.0 to R4.0). Fields were divided into 9 quadrates for a size of 0.4 ha or 20 quadrates for 4.0 ha. Twelve plants were chosen at random from each quadrate at each sampling date by removing the head and sealing it within a plastic bag. In the laboratory the total number of the adult weevils was counted on each head. The growth stage of each head was also recorded. Samples were taken at

3 to 6-day intervals and continued until >50% plants reached R5.7-R6.0 stage. The sampling period lasted 9 to 11 d.

The fields were also sampled for achene damage to determine the relationship between the adult weevil population and the subsequent achene damage. The estimation of achene damage differed between 1992 and 1993. Because most infested achenes contain a single larva (Oseto & Braness 1979), in 1992 we estimated the number of infested achenes per head by collecting larvae emerging from the sunflower heads in the fields (Oseto & Braness 1979) and recovering the larvae remaining in the achenes at harvest. Three plants were chosen at random from each of nine quadrates at plant stage R7.0 and they were bagged individually with larval collection bags. Larvae were collected weekly until harvest and counted in the laboratory. These sunflower heads were cut off and individually harvested. The number of unemerged larvae (those remaining in the achenes) for each head was estimated from total weight of achenes, number of achenes in 10 g sub-sample, and number of larvae recovered in 150 achenes. In 1993, an x-ray technique (Oseto & Korman 1986) was used to evaluate achene damage. When plants reached physiological maturity, twelve plants were selected randomly from each quadrate. The heads were dried for about a week at 41°C. The achenes were hand threshed, winnowed and weighed. To estimate the number of damaged achenes for each head, the total number of achenes and infestation percentage were determined. The number of achenes in a 20 g sub-sample was counted. From the 20 g sub-sample, 300 achenes were randomly selected and x-rayed to determine the number of infested achenes. The number of damaged achenes for each head was calculated from the total number of achenes and the infestation percentage.

In 1992, an additional 12 heads were harvested from each quadrate of the Minot field to determine weight and oil content loss in infested kernels. One hundred and fifty achenes were randomly selected from each head and dehulled. The damaged and undamaged kernels for each head were weighed, respectively, giving mean weight per kernel. The weight difference between the means of damaged and undamaged kernels gave weight loss per kernel. Damaged and undamaged kernels were pooled together respectively for oil content (%) analysis. Oil content was determined by using an Oxford 4000 NMR analyser (Oxford Analytical Instruments Limited, England). Kernel samples of about 35 ml each were dried in a laboratory oven at 65°C for 24 h before analysis. Ten samples each of damaged and undamaged kernels were analyzed. The oil content was reported based on kernel weight. The difference in means of oil content between damaged and undamaged kernels gave oil content loss. A statistical difference in oil content between damaged and undamaged kernels was examined using analysis of variance (SAS Institute 1987).

Results and Discussion

Relationship Between the Number of Adults and the Number of Damaged Achenes. In all three fields the density of the weevil population increased as plants developed from the bud stages (R2.0 to R4.0) to the anthesis stages (R5.0 to R6.0) (Table 1). If we ascribe the ultimate number of damaged achenes (or larval population) to the number of weevils sampled at different dates, we get different values. For example, the number of larvae per weevil that could be ascribed to the Minot, 4 August 1992 sample mean of 3.48 weevils per head was 58.92 (Table 1). At that time, most plants were in the bud stages. On 10 August, when most plants were in the anthesis stages the weevil population had increased and the number of larvae per weevil dropped to 29.50 (Table 1).

Insecticides are not directed against larvae because eggs and larvae are internal (Brewer 1991) and are not exposed to contact insecticides. Therefore, insecticide applications are designed to kill adults and prevent oviposition. The goal of adult weevil population sampling is to predict the ultimate achene damage (or size of the larval population) and the need for insecticide controls. Samples taken during the initial period of oviposition would be most predictive. Females begin to lay eggs on plants at the R5.4 stage and plants in the bud stages are unsuitable for oviposition (Oseto & Braness 1979). However, because there is a time delay between the taking of a sample and the application of controls sampling should be done before

the ideal plant stage (R5.4) for insecticide treatment (McBride et al. 1992). Because sunflower plants in fields do not develop uniformly weevil counts taken when most of plants are in the stages R5.0-5.3 are used to predict the subsequent number of damaged achenes (or larvae). Weevil counts on 10 August 1992 and 17 August 1993 at Minot were taken when most plants were in the stages R5.0-5.3 (see Table 1). The number of larvae ascribed to each weevil sampled was 29.50 in 1992 and 19.74 in 1993 at Minot. In the Prosper field, weevil counts were taken on 10 August when most plants were in the bud stages and again on 16 August when most plants were in the stages R5.7-6.0 (Table 1). The two weevil counts were taken either too early or too late. So, we used the mean of the weevil counts for both dates (0.29) to calculate the number of damaged achenes produced per weevil count, 31.40. The grand mean of the number of damaged achenes produced per weevil for the three fields is 26.88, which means that for every weevil sampled in plant stages R5.0-5.3, an average of 26.88 damaged achenes results if control measures are not taken. Because the sex ratio of the weevil population is reported to be 1:1 (Oseto & Braness 1980), the number of damaged achenes produced per a female must be doubled, i.e., 53.76. Actual female reproductive rate in the field conditions may be lower than the value of 53.76 because the weevil population may increase after plant stages R5.0-5.3. We found a weevil population increase after R5.0-5.3 stages in two of the three fields (Table 1). However, the increase was small. Apparently female reproductive rate in fields is higher than the value of 19.9 larvae per female which was used by Oseto and Braness (1980) to construct the EIL.

Damage. From 108 sunflower heads harvested at the Minot field (1992), 16,200 achenes were sampled and dehulled. The mean weight per damaged kernel was 27.66 ± 4.89 mg (n = 108). The mean weight per undamaged kernel was 37.53 ± 5.02 mg (n = 108). Thus, the mean weight loss per kernel was 9.86 ± 2.36 mg (n = 108), which is the amount of kernel consumed by a single larva. This single larval consumption is lower than the value Oseto & Braness (1980) used to calculate the EIL (12.8 mg).

The oil content for damaged kernels was 55.43 ± 0.29% and 58.77 ± 1.19% for undamaged kernels, showing significant reduction in oil content (Df = 1,18; $F = 74.31$; $P < 0.0001$). The 3.34% loss in oil content may result from a change of physiology in damaged kernels or frass left by larvae.

Development of Economic Injury Levels. We modified Calvin's (1985, 1988) cost/benefit (C/B) ratio formula to calculate the EIL for the red sunflower seed weevil. Calvin calculated the C/B ratio as:

$$C/B = \frac{(CC + AC) \times NA}{MV \times EY \times TPL \times PC} \qquad (1)$$

CC is the cost of chemicals (dollars per hectare), AC is the cost of application (dollars per hectare), NA is the number of applications, MV is the expected market value (dollars per kilogram), EY is the expected yield (kilograms per hectare), TPL is the total proportional yield reduction caused by a larval infestation, and PC is the expected proportional control from an insecticide application.

Oilseed sunflower growers are paid on the basis of yield and receive a premium or a penalty for sunflower with oil content above or below certain limits. Therefore, both yield and oil content losses due to larval infestation result in economic loss. Application of insecticides has two goals; to prevent yield loss (YL) and to prevent oil content loss (OL). Therefore, formula (1) can be rewritten as:

$$C/B = \frac{(CC + AC) \times NA}{YL + OL} \qquad (2)$$

The yield loss (YL) can be estimated based on the denominator of formula (1). The product of EY and TPL is actually the net yield loss (kilograms per hectare) due to larval consumption. In the case of the damage by the larvae of weevils, the net yield loss can be estimated by the product of weight loss per damaged achenes (WL), the number of damaged achenes per plant (ND) and plant population (PP). The ND is a product of the weevil counts (WC) in the plant

stages R5.0-5.3 and the resulting number of damaged achenes per weevil (DA). Therefore, the YL can be expressed as:

$$YL = MV \times WL \times WC \times DA \times PP \times PC \quad (3)$$

Oil content reduction varies with infestation percentage. We found a 3.34% oil content reduction for damaged kernels. However, achene oil content is used to determine the premium price. Oseto and Braness (1980) reported that mean oil content for undamaged achenes (0% infestation) is 43.80% and 32.74% for damaged achenes (100% infestation). We assume a linear relationship exists between infestation percentage and oil content. When the oil content is regressed against the infestation percentage using the above data (SAS Institute 1987), a slope of 11.06 is obtained. The slope means that for an increase of 10% infestation there will be a decrease of 1.106% oil content. A general rule for determination of the bonus price for oil is that for every one percent of oil content increase above a base value (usually 40%) there will be a 2% increase of the market value. Therefore, the oil content loss (OL) can be estimated using the following formula:

$$OL = EY \times \frac{WC \times DA}{NAP} \times PC \times 11.06 \times MV \times 0.02 \quad (4)$$

NAP is the average number of achenes per plant. The YL and OL in formula (2) are replaced by (3) and (4), resulting in the following formula:

$$C/B = \frac{(CC + AC) \times NA}{MV \times WL \times WC \times DA \times PP \times PC + EY \times \frac{WC \times DA}{NAP} \times PC \times 11.06 \times MV \times 0.02} \quad (5)$$

The number of weevil counts (WC) that results in a C/B ratio of 1 is the EIL value. If the numerator of the equation represents the total control cost (TC), formula (5) can be derived for EIL (WC):

$$EIL\ (WC) = \frac{TC}{1 \times MV \times DA \times PC\left(WL \times PP + EY \times \frac{1}{NAP} \times 11.06 \times 0.02\right)} \quad (6)$$

The market price of oilseed sunflower from 1981-1990 ranged from \$0.15/kg to \$0.29/kg (North Dakota Agricultural Statistics Service 1991). The current cost of one aerial application for registered insecticides is approximately \$18.53/ha. With two applications, the cost of control increases to \$36.06/ha. In a small plot insecticide trial, the control efficiency ranged from 91.3% to 99.5% larval reduction with two applications (Oseto & Braness 1980). We assume an 80% control efficiency for one insecticide application and 95% for two applications. Plant population also varies, ranging from 45,000/ha to 55,000/ha. From 1981 to 1990 in North Dakota, oil sunflower yields averaged 1261.6 kg/ha (North Dakota Agricultural Statistics Service 1991). The 348 plants sampled from Minot and Prosper in 1993 had a mean of 1400 achenes per plant.

Assuming either one or two spray applications, values for DA, PC, WL, EY, and NAP are constant, so equation 6 can be further derived:

For a single application, $$EIL_1 = \frac{TC}{MV \times 21.5\left(0.00986 \times \frac{PP}{1000} + 0.2\right)} \quad (7)$$

For two applications, $$EIL_2 = \frac{TC}{MV \times 25.54\left(0.00986 \times \frac{PP}{1000} + 0.2\right)} \quad (8)$$

The EIL values shown in Table 2 are calculated based on equations 7 and 8 using the proceeding information. The EIL is influenced by many factors as indicated in formula (6), especially the number of insecticide applications, market values and plant population. The number of insecticide applications determine the cost of control and control efficiency. Two applications of insecticide significantly increase the EIL values compared to one application, when other variables remain constant (Table 2). As indicated above, the market price fluctuated greatly in the 1980's. A Change in the market price results in an inversely proportional change in EIL values. For example, when the market price drops from \$0.30/kg (i.e., \$0.29/kg in 1983)

to $0.15/kg (i.e., 1986) the value of EIL is doubled (see Table 2). The plant population has much less impact on the EIL than the number of insecticide applications and the market value because the change of plant population is small.

The EIL values calculated using equations (7) and (8) are lower than the values based on Oseto and Braness' (1980) method (Table 2). The lower values of the EIL are attributed to the following reasons. First, we considered oil content loss in the calculation of the EIL, but they did not. Second, we used an average of 26.88 damaged achenes produced per weevil to estimate damage from weevil counts. They used an average of 19.9 larvae produced by a single female (approximately 10 damaged achenes produced per weevil count) to estimate the damage. Although higher cost of control and lower value of single larval consumption were used in our calculation, which lead to an increase of the values of EIL, their impact did not offset the inclusion of oil content loss and use of a higher value of damaged achenes produced per weevil count.

Our study specifies the use of weevil counts, when most of the plants are in the anthesis stages of R5.0-5.3, to make a treatment decision. This requires a precise and quick sampling procedure to estimate the weevil population density before the majority of the plants exceed the ideal stage for insecticide application. Our early study indicates an aggregated distribution of the adult weevils (Peng & Brewer 1994). The development of a sequential sampling program for the adult weevil is currently underway.

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Table 1. The relationship between the number of red sunflower seed weevil counts at different plant stages and the subsequent number of larvae collected or number of damaged achenes per head

Sampling date	Plant population structure (%)						Mean no. weevils per head	Mean no. larvae or damaged achenes per head	Mean no. larvae or damaged achenes per weevil
	R2.0	R3.0	R4.0	R5.0-5.3	R5.4-5.6	R5.7-6.0			
Minot, 1992									
4 August	7.4	53.7	40.0	0.9	--	--	3.48		58.92
10 August	--	4.6	13.0	36.1	29.6	16.7	6.95		29.50
13 August	--	--	2.8	18.5	19.4	59.4	6.46		31.74
Larval collection								205.04 ± 330.33	
Minot, 1993									
13 August	--	42.5	55.8	0.4	--	--	0.92		36.05
17 August	--	7.9	32.5	55.0	4.6	--	1.68		19.74
20 August	--	1.3	7.1	35.0	35.0	21.7	2.10		15.80
24 August	--	--	--	1.3	6.7	92.1	2.00		16.59
Damaged achenes								33.17 ± 43.78	
Prosper, 1993									
10 August	--	9.3	42.6	34.3	13.9	--	0.21		42.62
16 August	--	--	--	8.3	22.2	69.4	0.36		24.86
19 August	--	--	--	--	1.9	98.1	0.97		9.23
Damaged achenes								8.95 ± 12.12	

Table 2. Comparison of economic injury level values calculated using equations (7) and (8) and Oseto and Braness' (1980) method (in parenthesis) for the red sunflower seed weevil adult population when control cost, efficiency of control, plant population and market price are varied

Market price ($/kg)	Plant population 45,000/ha	50,000/ha	55,000/ha
	One application (TC = 18.53$/ha, PC = 80%)[a]		
0.15	8.9 (26.8)	8.3 (24.1)	7.7 (22.0)
0.20	6.7 (20.1)	6.2 (18.1)	5.8 (16.5)
0.25	5.4 (16.1)	5.0 (14.5)	4.6 (13.2)
0.30	4.5 (13.4)	4.1 (12.1)	3.9 (11.0)
	Two applications (TC = 36.06$/ha, PC = 95%)[a]		
0.15	14.5 (44.0)	13.6 (39.5)	12.7 (36.0)
0.20	11.0 (33.0)	10.2 (29.7)	9.5 (27.0)
0.25	8.8 (26.4)	8.2 (23.7)	7.6 (21.6)
0.30	7.3 (22.0)	6.8 (19.8)	6.3 (18.0)

[a] TC, total control cost; PC, expected proportional control.

Comparison of lab-determined deterrence with field cage deterrence to feeding by the sunflower beetle.

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Comparative analysis of feeding preferences for various populations of plants may disclose that some lines are less preferred than others. This difference of preference is a type of 'resistance' useful for protection of sunflower against pest species. Some types of preference may be more useful than others, since in the absence of a choice, some lines will be fed upon even if they are not preferred. For a type of preference that is not apparent except in the presence of a preferred line, cultural adjustments may need to be made. In this paper, we will show that a mixture of various preferred and non-preferred lines might be necessary to fully use this type of resistance successfully.

Methods

Various combinations of the two cultivars were planted in field cages 6' x 6' x 8', randomly placing sunflower in the rows and columns. A seven by seven matrix of plants spaced at 10 inch intervals was established, and 100 adult sunflower beetles were released in each cage. The ratios were 100%, 75%, 50% and 25% of each variety compared to the second. The 4th, 5th and 6th pairs of leaves were scored twice at 24h intervals, counting the apparent number of feeding initiation sites.

Results

From laboratory bioassay, we showed that in a group of eleven cultivars and hybrids evaluated against the hybrid 3311, some cultivars were significantly different in feeding preferences from IS 3311 (see Table 1). Line ST 365 was no different from IS 3311, but line RHA 274 was significantly different at $p<0.001$. From this, we assumed that RHA 274 and ST 365 would be significantly different in a field comparison against one another.

When no choice was offered caged beetles, the level of feeding was similar in both cultivars (see Table 2). In the experiment with 75% ST (the preferred line) the number of feeding initiation sites on ST were greater than those on the RHA plants (the less preferred line), and similar to the number of feeding initiations in the cage containing 100% ST. The RHA plants in this cage had fewer feeding sites than did plants in the cage with 100% RHA.

When the percentage of RHA (less preferred) was increased (to 50%), the feeding on RHA also increased to that of the no-choice controls (100% RHA) while feeding on the preferred ST increased to even more than that on its 100% ST controls. The amount of feeding on ST increased even more when the preferred line was 25% of

the total planting in a cage, while feeding on the RHA line also increased to greater than that in its 100% control.

In some combinations of preferred and less-preferred sunflower, we had higher or lower levels of feeding than in solid plantings of a single cultivar. What happened to the feeding rates overall is also of some importance to the success of this strategy. When we calculated the total numbers of feeding sites on all plants from the average numbers of holes and the number of those plants in the cage, we observed a trend of increasing amounts of feeding with decreasing amounts of the preferred line (Table 3). That is, the total feeding on the 75% ST cage was only slightly lower than the no-choice ST and RHA controls (100% plantings). However, the feeding in the cages with the lowest percentage of ST plants (25-50%) showed that the total number of holes increased with each decrease in percentage of non-preferred plants.

Conclusions.

It appears that when a cage contains the highest percentage of preferred plants, feeding pressure is relaxed on the least preferred plants. At 50% preferred plants, there is no benefit to the non-preferred plants. At less than 50% preferred plants, there appears to be an overall enhancement of feeding on the non-preferred as well as on the preferred plants.

Dual Choice Test: Leaf Areas Consumed (cm^2).

	Mean	SD	F	Mean	SD	F
	Experimental			IS 3311		
RHA 274	10.96	2.77	60.0*	4.63	1.31	3.36
ST 365	6.96	2.63	5.57	4.48	3.16	4.10

* $p>0.001$

Table 1

Feeding Sites per Leaf on Successive Days

Leaf Position:		4th pr	4th pr	5th pr	5th pr	6th pr	6th pr	
Assay / Day		2	3	5	6	8	11	Ave[1]
		Number of Feeding Sites Per Leaf						
75% ST	ST	2.86	4.49	4.71	6.51	2.66	4.51	4.6
	RHA	1.75	3.33	2.63	2.73	1.73	3.2	3.1
50% ST	ST	3.2	6.79	10.37	8.33	3.29	7.8	7.6
	RHA	3.32	6.36	7.92	5.08	3.2	4.7	5.4
25% ST	ST	3.18	8.08	8.91	11.08	3.92	7.9	9.0
	RHA	2.54	4.86	4.79	7.66	2.88	6.5	6.3
All ST		2.73	4.93	5.85	5.63	1.89	3.9	4.8
All RHA		2.31	5.65	3.83	4.39	2.88	5.5	5.2

1. Average scores from second day of 4th, 5th and 6th leaves.

Table 2.

Total Number of Feeding Sites Per Cage

Ratio of Plants			Sites fed upon
All ST			480
75% ST	ST	4.6 X 75	345
	RHA	3.1 X 25	77
	Total		422
50% ST	ST	7.6 X 50	380
	RHA	5.4 X 25	270
	Total		650
25% ST	ST	9.0 X 25	225
	RHA	6.3 X 75	473
	Total		698
All RHA			520

Table 3

Patterns of Phenylpropanoid Expression in Sunflower Leaves and Deterrence of Sunflower Beetles to Leaf Feeding

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Phenylpropanoids are a large group of plant compounds with the basic structure of a C_6 phenyl ring and a C_3 propane chain. The key reaction leading to the synthesis of these compounds is that catalyzed by phenylalanine ammonia lyase (PAL), in which phenylalanine is converted to cinnamic acid. Subsequent reactions produce a wide variety of phenylpropanoid compounds with a wide range of functions. Included in this group are flower pigments, antibiotics, UV protectants, and insect deterrent compounds, as well as signal molecules important in plant-microbe interactions and polymeric constituents of cell wall structures. (1,3,4)

This study involved an attempt to identify those phenylpropanoids with feeding detterent properties in sunflower leaf tissue by coorelating feeding response with phenylpropanoid patterns. Sunflower accessions that varied in their resistance to sunflower beetle feeding were tested.

Materials and Methods

Plant Material. The first group tested consisted of wild *H. annuus* accessions chosen on the basis of damage by larval sunflower beetles. Two of the accessions had high numbers of larvae feeding on them, (according to data from the Plant Introduction Station, Ames, IA) and the other 8 accessions had relatively low numbers of larval feeding, indicating possible resistance mechanisms.

	Accession #	Origin	
I	PI 386230	USSR	High SFB feeding
II	PI 162675	Argentina	
III	PI 170389	Turkey	
IV	PI 175726	Turkey	High SFB feeding
V	PI 181881	Syria	
VI	PI 226466	Iran	
VII	PI 408726	France	
VIII	PI 413034	US/Nebraska	
IX	PI 431513	Romania	
XI	PI 431535	Yugoslavia	

Seeds were pre-germinated and seedlings transplanted to 4-in. square pots in the greenhouse. Leaves were sampled after 4-5 weeks for phenylpropanoid and bioassay studies.

The second group tested consisted of inbred lines obtained from USDA, Fargo, ND, and several hybrids. These were germinated and grown in the same manner as the first group.

A	ARG 420
B	RES 834
C	ANO 1509
D	ST 316
E	IS 3311
F	RHA 274
G	HIR 1734
I	HA 303
J	RHA 365
L	HA 89

Phenylpropanoid Assay. The method used was that of T. Graham (2). Leaf disks of 1-cm diameter were obtained from the 4th leaf pairs, and this tissue was homogenized in 80% ethanol to a concentration of 250 mg/ml, and centrifuged. The supernatant was applied to an HPLC column, and phenylpropanoids eluted on a Lichrosorb C18, 10µ reverse-phase column. The mobile phase consisted of a 0 - 55% acetonitrile/pH 3 water linear gradient over 25 min, followed by a step increase to 100% acetonitrile to remove tightly bound compounds. Flow rate was 1.5 ml/min, temperature 30 C, and peaks were detected on a UV/VIS spectrophototmeter at a wavelength of 254 nm. A mixture of 6 known phenylpropanoid compounds was run as a standard. Selected unknown sample peaks were quantitated by comparison with the chlorogenic acid standard.

Feeding Choice Bioassays. A dual choice feeding test with sunflower beetle (*Zygograma exclamationis,* L.) adults was conducted using the test accession and a "standard" hybrid (Interstate 3311). Leaves (4th pair) were cut at the base of the petiole and stems placed in small vials of distilled water. Two leaves of each type were placed in an aluminum pie plate arena with moist filter paper lining the bottom. Eight sunflower beetle adults (starved for 24 hrs before testing) were added, and the arena covered with a clear plastic lid. After 24 hrs, the results were tabulated by noting the number of feeding sites per leaf. A feeding score was obtained for each arena by using the formula "2T/T+C", where T= # of feeding sites on the test leaves and C= # of feeding sites on the control leaves. A high score (close to 2) indicates a preference for the test leaves, wheras a low score (close to 0) would indicate that the test leaves are detterent.

Bioassays for the second group of plants were performed in a similar manner, except in the reading of the results. After feeding, the leaves were read on a leaf area meter to obtain the final leaf area. To get an estimate of the inital leaf area, the leaves were then exposed on photographic film, and the images cut out according to their surmised initial shape. These images were then read in the leaf area meter. Final areas were subtracted from initial areas to obtain areas eaten. Feeding scores were obtained using the formula "2T/T+C", where T= the area eaten on the test leaves, and C= the area eaten on the control leaves.

Results

Refer to Table 1 for the complete phenylpropanoid chromatogram results for the first group of wild *H. annuus* accessions tested. To identify those phenylpropanoid peaks with possible feeding deterrent properties, the accessions having the lowest feeding scores were selected (I, IV, and VII). The phenylpropanoid peaks that were higher in these accessions, relative to the other accessions were then chosen. An X-Y scatter plot was generated with feeding scores of all accessions on the y axis, and their corresponding phenylpropanoid concentrations on the x axis. A graph was generated for each of the chosen phenylpropanoid peaks, and simple regression lines were drawn. The slope of

this line gives an indication of the relationship between the 2 variables. An negative slope (going downward from left to right) would indicate a possible feeding deterrent compound (attraction is decreasing as phenylpropanoid concentration is increasing). A positive slope, however, might indicate a feeding attractant. Figure 1 shows an example of a deterrent, and an attractant compound.

Table 2 shows all the phenylpropanoid peaks quantitated in the second group of sunflower lines. Lines C, G, and I had the lowest feeding scores when tested against the standard cultivar Interstate 3311. The same method was used to generate graphs, and Figure 2 shows a deterrent compound, and an attractant compound for this group.

Another analysis strategy involved first identifying all possible detterent and attractant compounds (based on the above information). The total concentration of all deterrents was then divided by the total concentration of all attractants to arrive at a D/A ratio, and this ratio was plotted (on the x axis) against feeding scores (on the y axis). We would hope to see a negative slope here, since that would indicate that an increase in concentration of deterrent compounds coorelates with decreased insect feeding (Figure 3). For the wild sunflower accessions, peaks at 10.55, 1922, 19.78, and 26.3 min. were initially chosen as deterrents. Attractants were tentatively identified at 12.5, 14.57, 17.07, and 22.85 min. When the D/A ratio was plotted against the feeding scores, a negative slope was obtained, so the peaks were probably identified correctly as deterrents and attractants (Figure 3).

For the next group of sunflower lines, deterrents were 19.67, 20.78, and 21.4 min. peaks, and the attractants at 13.95, 16.35, 16.92, and 17.4 min. With these choices, however, the graph of D/A vs. feeding score showed a completely flat slope (Figure 3). This may indicate that deterrents and attractants were not identified correctly. The analysis of this data will have to be worked up more extensively to reach conclusions about deterrent compounds, and other analysis schemes are currently being evaluated.

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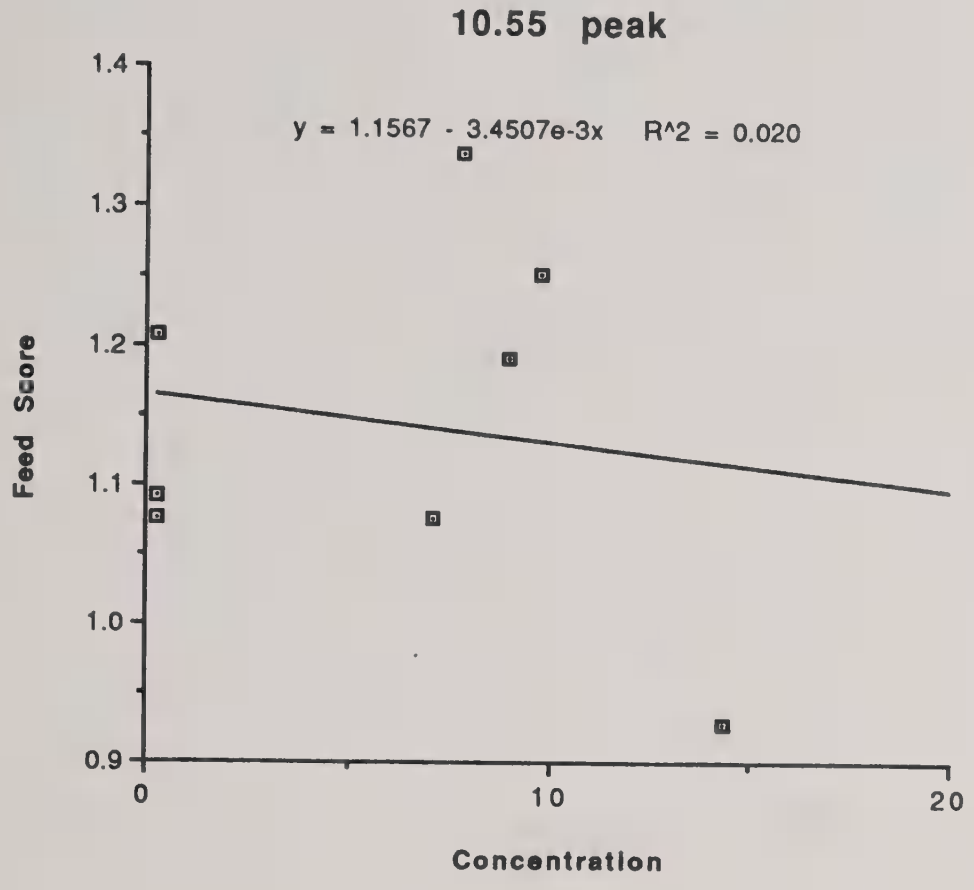
		Phenylpropanoid Peaks - Retention Time (min)															
Accession	Feed Score	10.55	10.8	12.5	13.3	14.57	15.64	16.26	16.52	17.07	17.85	19.22	19.78	20.4	21.35	22.85	25.14	26.3
I	0.919	14.1	52.4	1222.7	13.7	15.9	17.5	93.6	111.2	1475.0	21.0	62.0	88.0	40.4	854.3	48.2	22.5	46.7
II	1.200	0.0	29.3	2811.7	24.0	0.0	21.6	59.8	52.0	1221.4	0.0	65.4	60.4	31.0	318.1	38.4	0.0	0.0
III	1.244	9.5	57.2	5825.3	133.6	13.5	55.9	90.5	168.3	4366.5	278.9	54.2	47.3	37.2	212.0	47.0	18.7	0.0
IV	1.067	0.0	22.5	559.2	0.0	0.0	0.0	49.6	55.4	667.0	12.8	69.5	84.3	73.6	202.4	62.7	22.9	0.0
V	1.067	6.8	52.1	1805.9	62.9	17.0	21.9	102.9	182.1	1935.1	138.6	64.5	142.6	96.5	268.4	59.7	22.5	11.2
VI	1.329	7.5	45.5	3655.0	97.2	20.0	12.1	73.7	152.6	2624.4	98.5	65.0	60.0	91.1	312.5	77.6	25.9	0.0
VII	1.084	0.0	0.0	186.9	11.8	0.0	0.0	24.5	10.8	182.1	0.0	155.5	137.7	37.8	127.4	10.5	0.0	0.0
IX	1.183	8.7	38.6	2808.5	26.5	15.4	28.6	155.9	197.3	2416.9	63.1	151.7	126.6	57.1	231.8	26.5	7.6	0.0
XI	1.420	31.1	63.3	3860.4	81.4	24.5	25.5	128.7	0.0	2569.9	82.9	159.4	144.3	32.8	372.9	17.2	11.1	0.0
3311		12.4	0.0	15.4	0.0	25.3	0.0	9.1	0.0	10.7	0.0	163.3	190.7	117.9	334.7	29.6	7.3	0.0

Table 1. Phenylpropanoid concentrations (µg CA-equivalent/µg fresh wt) and feeding scores for sunflower accessions in Group 1.

		Phenylpropanoid Peaks - Retention Time (min)															
Line	Feed Score	10.68	11.15	11.73	12.39	12.59	13.15	13.39	13.68	13.95	14.31	14.68	15.69	15.98	16.35	16.52	16.92	17.40
A	1.467	38.1	28.7	18.6	17.2	36.3	86.3	66.7	34.6	29.4	21.5	19.5	20.7	31.5	55.8	23.6	81.8	491.5
B	1.445	16.5	11.3	9.7	3.4	27.1	47.2	55.1	19.7	7.4	22.4	7.7	15.8	10.0	63.3	0.0	61.5	444.0
C	1.077	10.8	3.2	15.4	5.9	28.6	3.4	17.5	0.0	0.0	35.9	14.9	14.3	8.3	29.1	9.8	7.5	51.8
D	1.293	13.3	34.9	28.8	0.0	5.7	130.5	27.0	0.0	0.0	0.0	6.3	9.9	0.0	0.0	7.3	0.0	4.7
E	1.202	18.8	24.4	73.7	5.6	8.8	31.9	56.1	34.6	7.8	5.3	13.0	10.6	5.4	9.5	0.0	5.4	21.3
F	1.405	36.1	18.7	2.8	0.0	44.0	57.6	0.0	4.3	4.8	13.7	20.3	9.3	7.2	18.2	0.0	14.6	61.8
G	1.165	15.7	4.0	25.4	7.1	47.8	27.1	16.3	17.0	20.7	21.7	13.2	39.3	70.7	35.2	0.0	42.5	42.5
I	0.902	13.1	2.2	8.2	3.2	32.5	27.5	5.8	7.6	13.2	3.4	3.4	2.5	5.1	0.0	5.7	21.5	24.4
J	1.212	0.0	0.0	12.4	1.0	7.6	21.9	0.0	0.0	0.0	2.1	0.0	9.6	1.1	2.4	1.6	2.4	5.4
L	1.403	14.6	24.0	34.0	3.5	9.7	18.3	0.0	179.4	6.3	2.1	7.3	4.8	0.0	0.0	4.1	4.1	15.7

Line	19.31	19.67	20.17	20.51	20.78	21.40	22.89	24.09	25.06	29.30
A	125.5	192.0	35.7	71.7	32.9	44.4	31.3	4.1	3.1	36.3
B	53.4	91.3	0.0	21.4	1.6	46.2	23.3	4.0	5.6	130.5
C	86.5	166.0	22.3	99.9	23.4	176.6	31.2	4.0	4.4	70.5
D	18.6	15.3	0.0	65.7	5.1	598.1	65.9	2.7	12.7	24.1
E	14.8	12.4	0.0	9.4	0.0	118.1	9.6	4.0	4.2	26.6
F	135.2	217.9	70.6	26.2	34.5	171.9	0.0	5.4	0.0	54.5
G	2080.3	0.0	60.4	184.5	0.0	179.7	30.0	26.4	2.3	33.9
I	129.4	270.6	0.0	45.7	43.9	254.1	23.2	5.9	3.2	16.7
J	116.8	241.2	0.0	20.8	23.9	7.2	0.0	5.4	0.0	11.5
L	6.5	8.3	3.5	53.9	18.9	88.3	68.5	8.1	12.2	34.8

Table 2. Phenylpropanoid concentrations (µg CA-equivalent/µg fresh wt) and feeding scores for sunflower accessions in Group 2.



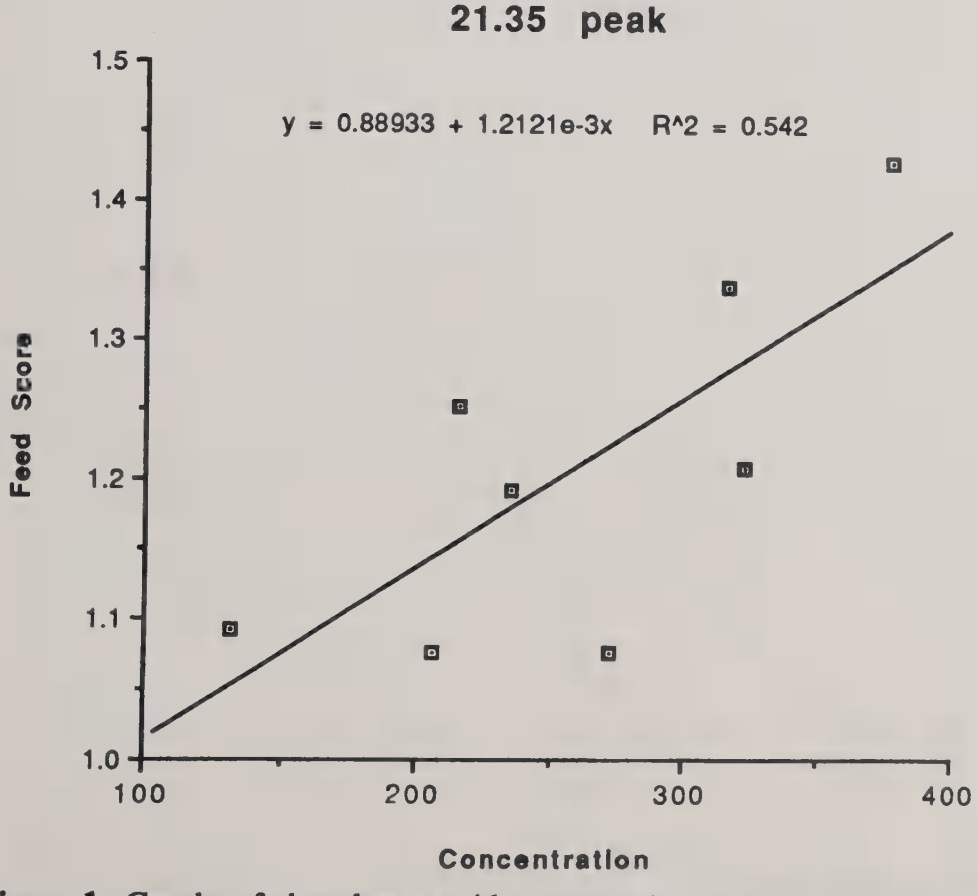
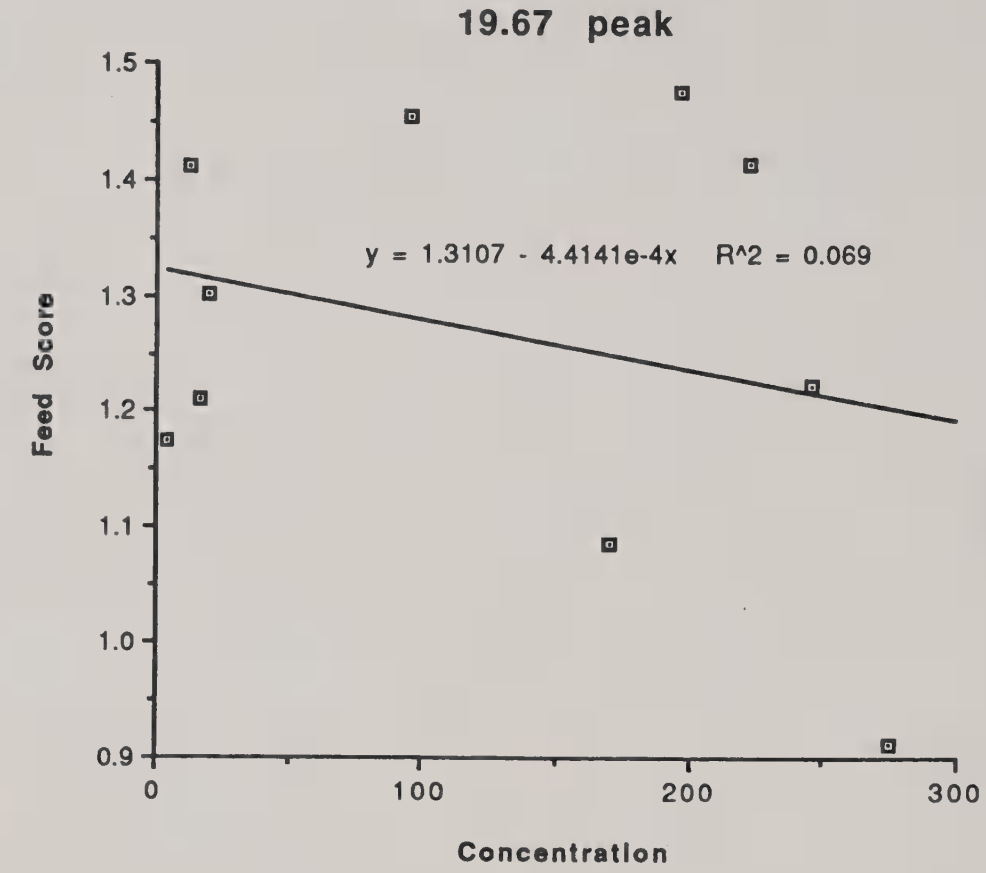


Figure 1. Graphs of phenylpropanoid concentration vs. feeding score for some representative peaks in the wild *H. annuus* accessions.



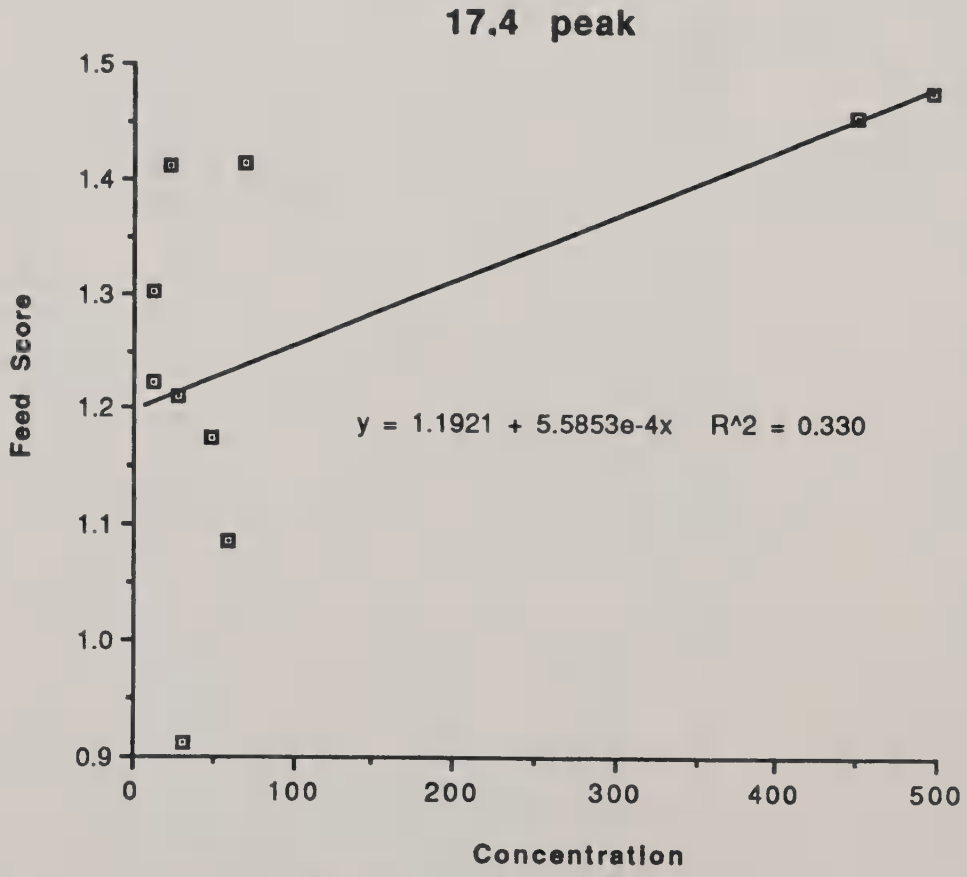
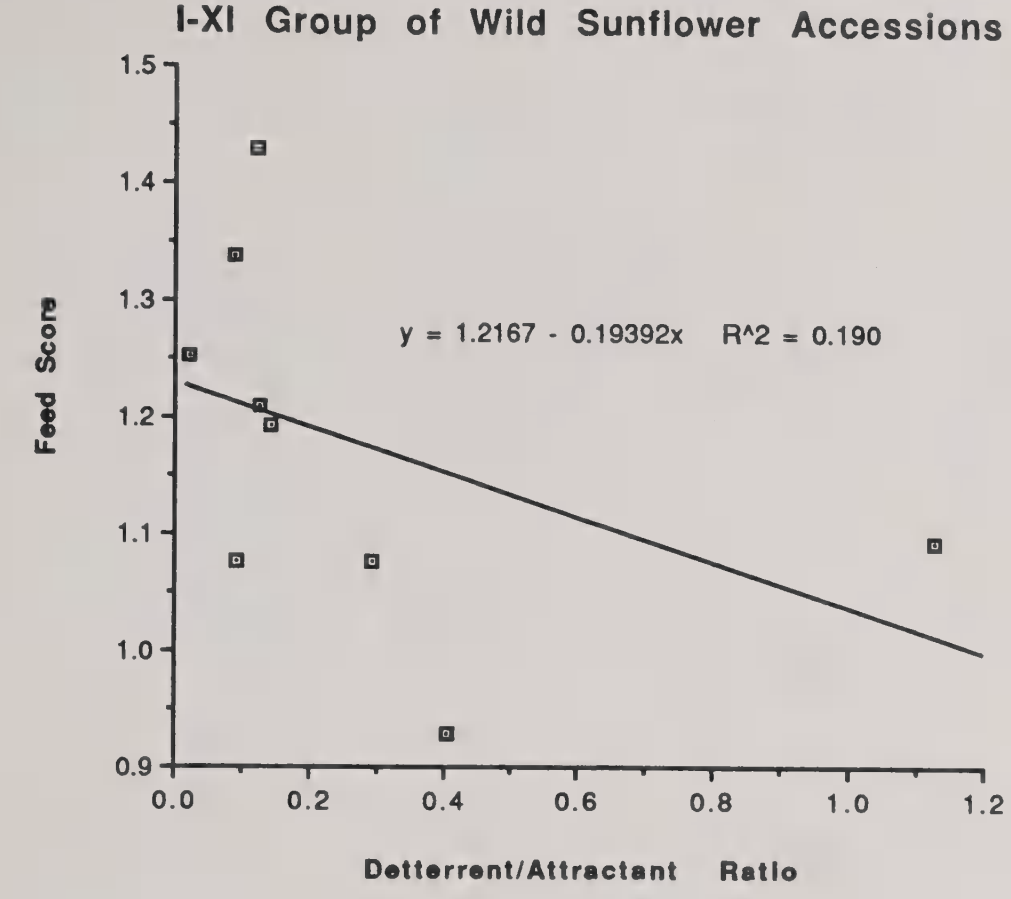


Figure 2. Graphs of phenylpropanoid concentration vs. feeding score for some representative peaks in the cultivated sunflower lines tested.



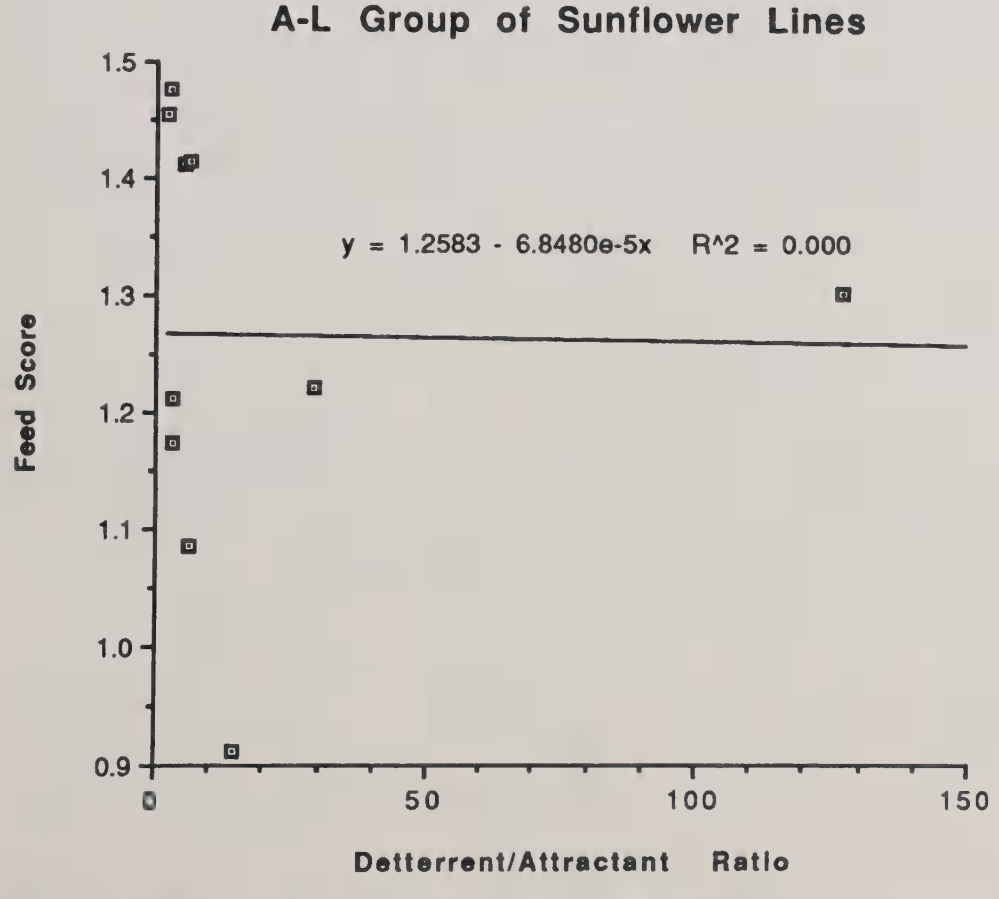


Figure 3. Graphs of deterrent/attractant ratios vs. feeding score for the wild *H. annuus* accesions (top) and other sunflower lines (bottom).

North Dakota Aerial Applicators Surveys 1983 - 1993

DAVID R. NELSON

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NORTH DAKOTA AERIAL SPRAYING QUESTIONNAIRES Estimated Acres Sprayed by Aerial Applicators for Selected Pests											
CROP/PEST	1983	1984	1985	1986	1987	1988	1989	1990	1991	1992	1993
% reporting	37	32	37	32	52	54	54	48	50	40	35
Head Moth	1,081	5,313	0	0	2,500	0	0	250	0	0	0
Banded Moth	946	20,359	3,027	2,344	1,538	0	0	0	0	0	0
Seed Weevils	329,703	673,703	627,997	554,744	588,162	354,894	1,042,015	572,352	1,075,694	411,428	129,508
Sunflower Beetle	790,411	1,148,400	293,235	235,522	66,373	42,128	4,546	3,333	19,810	28,673	144,350
Stem Weevil	4,324	112,106	45,381	43,728	45,165	9,630	44,704	504	105,240	13,675	6,433
Grasshoppers	23,489	51,372	54,454	31,713	6,802	4,381	37,700	13,356	86,516	7,338	0
GH + SW	4,803	0	0	3,125	14,735	1,852	20,983	32,029	50,994	0	0
GH + ST W	0	0	0	3,125	0	0	0	1,250	16,000	2,500	0
GH + SB	0	21,078	0	6,250	0	0	0	0	0	0	0
B + SEED W	0	0	0	0	0	0	0	0	0	0	16,740
B + GH + STEM	0	0	0	938	0	0	0	0	0	0	1,430
Complex	110,811	143,050	164,054	20,469	0	0	0	0	0	0	0
Cutworms	1,405	0	0	0	0	0	0	0	2,848	385	889
TOTAL	1,266,973	2,175,381	1,188,149	901,956	725,275	412,885	1,149,948	623,075	1,357,102	463,998	299,351

PROPORTION OF TREATED ACRES SPRAYED FOR EACH PEST
AERIAL APPLICATOR SURVEYS 1983 – 1993

NORTHWEST DISTRICT					
YR	Beet	SW	StW	GH	BM
1983	0.91	0.00	0.00	0.09	0.00
1984	0.56	0.00	0.00	0.44	0.00
1985	0.18	0.00	0.00	0.82	0.00
1986	0.00	0.62	0.00	0.38	0.00
1987	0.00	0.91	0.00	0.09	0.00
1988	0.00	0.99	0.00	0.01	0.00
1989	0.00	1.00	0.00	0.00	0.00
1990	0.00	0.83	0.00	0.17	0.00
1991	0.00	0.83	0.06	0.11	0.00
1992	0.20	0.78	0.01	0.01	0.00
1993					

NORTH CENTRAL DISTRICT					
YR	Beet	SW	StW	GH	BM
1983	0.94	0.02	0.00	0.04	0.00
1984	0.94	0.01	0.00	0.02	0.03
1985	0.65	0.06	0.01	0.27	0.02
1986	0.55	0.35	0.00	0.10	0.00
1987	0.23	0.75	0.00	0.00	0.01
1988	0.25	0.68	0.00	0.06	0.00
1989	0.00	0.97	0.00	0.03	0.00
1990	0.03	0.91	0.01	0.05	0.00
1991	0.01	0.85	0.00	0.14	0.00
1992	0.13	0.82	0.01	0.03	0.00
1993	0.48	0.52	0.00	0.00	0.00

NORTHEAST DISTRICT					
YR	Beet	SW	StW	GH	BM
1983	0.97	0.03	0.00	0.00	0.00
1984	0.83	0.12	0.03	0.00	0.02
1985	0.83	0.15	0.00	0.01	0.01
1986	0.57	0.40	0.00	0.00	0.03
1987	0.64	0.35	0.00	0.00	0.01
1988	0.54	0.46	0.00	0.01	0.00
1989	0.06	0.93	0.00	0.01	0.00
1990	0.01	0.98	0.00	0.01	0.00
1991	0.14	0.83	0.00	0.04	0.00
1992	0.31	0.69	0.00	0.01	0.00
1993	0.81	0.19	0.00	0.00	0.00

WEST CENTRAL DISTRICT					
YR	Beet	SW	StW	GH	BM
1983	0.00	0.00	0.00	1.00	0.00
1984	0.55	0.24	0.04	0.17	0.00
1985					
1986	0.00	1.00	0.00	0.00	0.00
1987	0.00	1.00	0.00	0.00	0.00
1988	0.00	1.00	0.00	0.00	0.00
1989	0.00	1.00	0.00	0.00	0.00
1990	0.00	0.97	0.00	0.03	0.00
1991	0.00	0.92	0.00	0.08	0.00
1992	0.00	0.93	0.00	0.07	0.00
1993	0.00	1.00	0.00	0.00	0.00

CENTRAL DISTRICT					
YR	Beet	SW	StW	GH	BM
1983	0.84	0.16	0.00	0.00	0.00
1984	0.59	0.28	0.13	0.00	0.00
1985	0.23	0.73	0.04	0.00	0.00
1986	0.43	0.43	0.08	0.05	0.00
1987	0.04	0.80	0.15	0.01	0.00
1988	0.07	0.87	0.05	0.01	0.00
1989	0.00	0.84	0.12	0.04	0.00
1990	0.00	1.00	0.00	0.00	0.00
1991	0.01	0.80	0.11	0.08	0.00
1992	0.04	0.87	0.07	0.02	0.00
1993	0.49	0.50	0.01	0.00	0.00

EAST CENTRAL DISTRICT					
YR	Beet	SW	StW	GH	BM
1983	0.18	0.82	0.00	0.01	0.00
1984	0.33	0.61	0.06	0.00	0.00
1985	0.11	0.73	0.17	0.00	0.00
1986	0.17	0.75	0.08	0.00	0.00
1987	0.04	0.88	0.06	0.02	0.00
1988	0.04	0.93	0.02	0.01	0.00
1989	0.00	0.93	0.00	0.07	0.00
1990	0.00	0.97	0.00	0.03	0.00
1991	0.00	0.82	0.13	0.05	0.00
1992	0.00	0.99	0.00	0.01	0.00
1993	0.30	0.70	0.00	0.00	0.00

SOUTHWEST DISTRICT					
YR	Beet	SW	StW	GH	BM
1983	0.00	0.00	0.25	0.75	0.00
1984	0.00	0.84	0.00	0.16	0.00
1985	0.00	0.85	0.00	0.15	0.00
1986	0.00	0.69	0.00	0.31	0.00
1987	0.00	0.97	0.00	0.03	0.00
1988	0.00	1.00	0.00	0.00	0.00
1989	0.00	1.00	0.00	0.00	0.00
1990	0.00	1.00	0.00	0.00	0.00
1991	0.00	0.90	0.00	0.10	0.00
1992	1.00	0.00	0.00	0.00	0.00
1993					

SOUTHCENTRAL DISTRICT					
	BEETLE	SEEDW	STEMW	GH	BAND
1983	0.00	0.17	0.00	0.83	0.00
1984	0.41	0.05	0.00	0.54	0.00
1985	0.00	0.83	0.06	0.11	0.00
1986	0.00	0.97	0.00	0.03	0.00
1987	0.00	1.00	0.00	0.00	0.00
1988					
1989	0.00	0.99	0.00	0.01	0.00
1990	0.00	1.00	0.00	0.00	0.00
1991	0.00	1.00	0.00	0.00	0.00
1992	0.00	1.00	0.00	0.00	0.00
1993	0.00	1.00	0.00	0.00	0.00

SOUTHEAST DISTRICT					
YR	Beet	SW	StW	GH	BM
1983	0.00	1.00	0.00	0.00	0.00
1984	0.27	0.66	0.06	0.00	0.00
1985	0.01	0.97	0.02	0.00	0.00
1986	0.09	0.87	0.04	0.01	0.00
1987	0.01	0.97	0.02	0.00	0.00
1988	0.01	0.98	0.00	0.01	0.00
1989	0.00	0.97	0.01	0.02	0.00
1990	0.00	0.96	0.00	0.04	0.00
1991	0.00	0.96	0.04	0.00	0.00
1992	0.00	0.96	0.03	0.00	0.00
1993	0.34	0.51	0.15	0.00	0.00

Sunflower insect pest situation during 1993 and prospects for 1994:

Colorado - Sue Blodgett, Stan Pilcher, Frank Peairs
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Completion of an oilseed processing plant in Goodland, Kansas has resulted in an increased acreage of oilseed sunflowers in eastern Colorado. Key insect pests of sunflower in Colorado are the larval stages of the sunflower moth, *Homeosoma electellum*, and the banded sunflower moth, *Cochylis hospes*, and the sunflower seed weevils, *Smicronyx fulvus* and *S. sordidus*. Native sunflower and other related native/resident plant species provide a constant source of potential insect pests for cultivated sunflowers.

Banded sunflower moth: Large numbers of banded sunflower moths were collected from pheromone traps (peak moth catches ranged from 20 - 40 moths per night) in most locations. Traps catches were much greater than anticipated for this insect. However, very few larvae were detected in sunflower heads and there was no relationship between the pheromone trap catches obtained and larvae per 10 heads. Although adult moths seemed to be abundant, our data indicated that high pheromone trap catches did not coorespond to high larval populations.

Sunflower moth: Pheromone trap catches of sunflower moths were very low (less than 1 moth per night) in all locations. Although pheromone trap catches were low, moth flight seemed to be correlated with larval populations in the sunflower heads. There was a stronger relationship between sunflower moths per jug trap per night and larval populations per 10 sunflower heads at the south field edge ($r^2 = 0.85$) than at the north field edge ($r^2=0.10$) for the sunflower moth.

Pheromone trap design and placement: Both wing traps (Scentry Inc.) and jug traps (1 gallon milk containers) baited with sunflower moth (Scentry Inc.) and banded sunflower moth (L. Bjostad, Colorado State University) pheromones were used to evaluate the effect of trap design on catch efficiency. Both trap types caught moths at approximately the same rate. More moths were caught in traps placed at the south field edge than the north field edge. Pheromone traps placed on south field edges showed a strong relationship between moth catches and larval populations, suggesting that trap placement was important.

Regional project for 1994:

Kansas State University: Gerald Wilde

University of Nebraska: Gary Hein

Colorado State University: Stan Pilcher, Frank Peairs

Preliminary data collected in eastern Colorado during the 1993 field season indicated that while large numbers of banded sunflower moths were present (detected in pheromone traps), larvae were found in very low numbers suggesting that wild sunflowers species may be acting as a reservoir for the pest species. A regional study proposed for 1994 will include determining the relationship between pheromone trap catches of banded sunflower moth and sunflower moth and larval infestations in untreated sunflower fields. Similar plot designs will be used in each location to determine if pheromone trap catches can be used as a reliable method for determining treatment timing of banded and sunflower moths. Wild sunflower plants at the same growth stage as the field trial will be inspected for immature stages of target insect species.